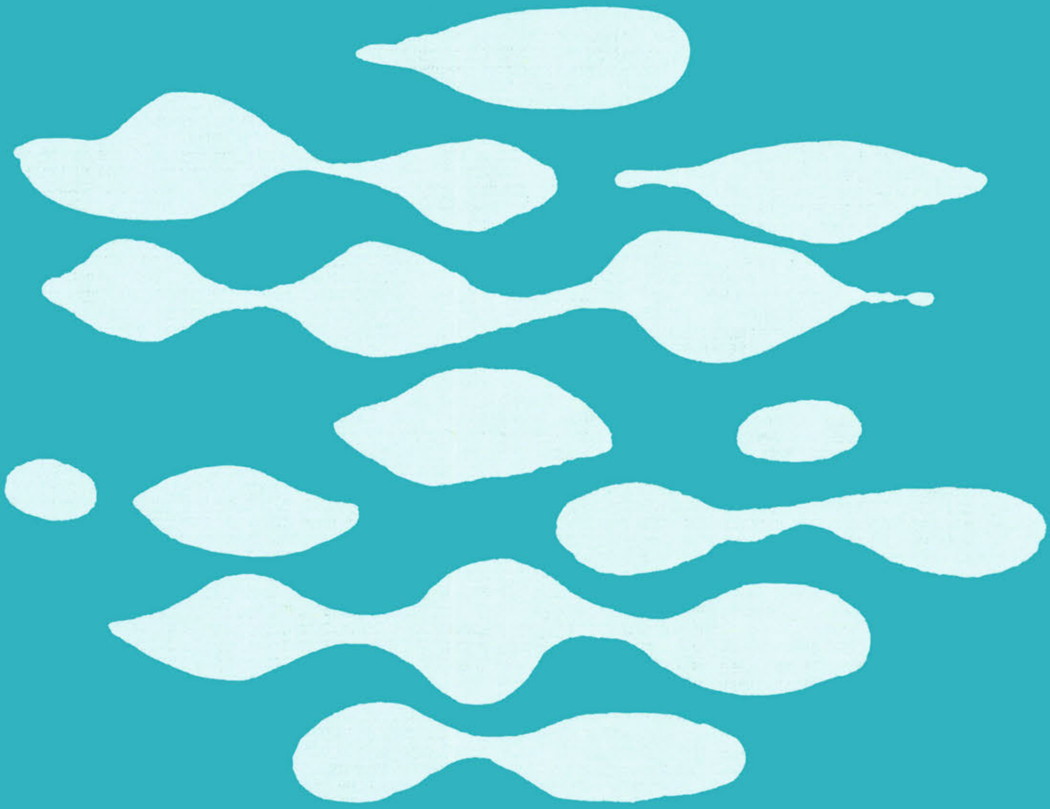
DEVELOPMENTS IN HYDROBIOLOGY

Nutrients and Eutrophication in Estuaries and Coastal Waters

edited by
Emma Orive, M. Elliott,
and V.N. de Jonge



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Nutrients and Eutrophication in Estuaries and Coastal Waters

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Nutrients and Eutrophication in Estuaries and Coastal Waters

Proceedings of the 31st Symposium of the Estuarine and Coastal Sciences Association (ECSA), held in Bilbao, Spain, 3–7 July 2000

Edited by

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Preface

The 31st annual international symposium of the Estuarine and Coastal Sciences Association (ECSA) was held in Bilbao, Basque Country, Spain, in July 2000. The symposium was organized by the University of the Basque Country, the Estuarine and Coastal Sciences Association (ECSA) and the Bilbao–Bizkaia Water Consortium. It was sponsored by the Basque Government (for County Planning, Housing and Environment, for Education, Universities and Research and for Agriculture and Fisheries), the Bilbao–Bizkaia Water Consortium, the County Council of Biscay, PETRONOR, the University of the Basque Country and the Spanish Government for Science and Technology. The organizers thank each of these organizations for their support in making the symposium successful.

The Symposium was planned to provide a forum for on-going research on nutrient and organic matter inputs and distribution in coastal ecosystems, and their effects on the geo-chemistry of the system and biological community structure. The discussions considered the possibilities and the ability for the recovery of systems to a trophic state that is of benefit to man and nature. The understanding of the causes and consequences of nutrients added to estuaries and coastal waters is important both for nature conservation and for the socio-economic aspects and uses of these areas. Whereas point-source pollution has been well-studied for several decades, the understanding of problems caused by diffuse pollution, such as nutrients from agricultural run-off, has received less attention until recently. Such diffuse pollution is harder to assess, quantify, study and control, features which contribute to its only recent attention by environmental managers and politicians. In addition to those inputs from land-based diffuse sources, organic matter and nutrients emanate from many other sources, from point sources in urban and industrialised areas, to atmospheric inputs. The effects of such inputs can be regarded as a set of symptoms of ecosystem pathology which indicate that the aquatic environment is undergoing undesirable change.

The papers presented at the symposium and those published in this volume reflect the range of studies currently undertaken and related to nutrients and potential or actual eutrophication. They represent the DPSIR approach in which the Drivers of change occur, i.e. the wide scale causes such as discharge and farming practices. Secondly, the Pressures on the system, such as particular inputs of nitrogen and phosphorous compounds. Thirdly, the Status of the estuarine and coastal systems as a reflection of changes brought about by such causes. Fourthly, the impacts such as the levels of contamination, hypernutrification, eutrophication and change at all levels of the biological system. Finally, the studies mention the Responses that can be made by Man, using management, legislative and administrative tools, to such drivers and pressures.

It is axiomatic that in order to understand the consequences of nutrients within such estuarine and coastal systems, it is necessary to understand both the physical and chemical relationships and features. This includes not just the hydrodynamics of an area, which will dictate the time with which nutrients are available to uptake by the biota and thus the start of any undesirable consequences, but also the behaviour of the nutrients. The latter dictates that a good knowledge is required of the transformations of nitrogen and phosphorous compounds, of the utilisation of organic matter and the links to carbon and silicon cycles. The proceedings here indicate that studies on these features are required as a precursor to understanding and then predicting the biological repercussions of nutrient additions. Again, it is only when undesirable consequences of such additions occur that the general public, politicians and environmental managers become aware of diffuse and nutrient pollution. Those undesirable consequences create the set of symptoms known as eutrophication which has far reaching repercussions for all users and uses of the marine and estuarine waters.

Above all, the symposium and this resulting set of papers indicate the need for a multi-disciplinary and cross-disciplinary approach to the study of nutrients and eutrophication in dynamic systems such as estuaries and coasts. Hence that study calls for hydrographers, chemists and biologists to link with environmental managers, social scientists and environmental economists and lawyers.

The Estuarine and Coastal Sciences Association (ECSA)

The Estuarine and Coastal Sciences Association (ECSA) is a direct continuation of the Estuarine and Brackish-Water Sciences Association /EBSA), founded in 1971. The Association is the major European focus for the communication of research and scholarship in estuarine science. Membership is open to all who are interested in estuarine and coastal marine science, whether in Europe or further afield. The association holds local meetings and annual symposia both within Europe and further afield, it produces proceedings of symposia and other publications as well as having its newsletter the Bulletin. ECSA has an associated journal "Estuarine, Coastal and Shelf Science", under the guidance of the Association's Editor Dr Donald McLusky, which is available at greatly reduced rates to members. ECSA has links to the Estuarine Research Federation in the US and thus contacts with the wider global estuarine and coastal community. Details of the Association's activities can be found on the ECSA website http://www.ecsa.ac.uk and membership and other enquiries should be directed to: Dr Trevor Telfer, Institute of Aquaculture, University of Stirling, Stirling, FK9 4LA, U.K.

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Causes, historical development, effects and future challenges of a common environmental problem: eutrophication

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Abstract

The impact of the presence of us humans as well as our activities to nature has led to over-exploitation of natural resources and to dramatic changes in land-use including the use of artificial fertilizers contributes to the deterioration of the natural environment. The population density, industrial processes and the use of fertilizers are the main causes for the eutrophication of river systems, estuaries and seas. There are several ways to determine the actual nutrient levels back to the 1950s, back to 1900 or even back to 1800. Available data indicate that the natural background concentrations of nutrients (pristine conditions or the period prior to the widespread use of artificial fertilizers and detergents) were dramatically lower than today. Available time series on chemical and biological data collected from different parts of the world show the (sometimes tremendous) increase in nutrient levels and the related productivity. The same time series, however, also show the decreases in values when measures were taken to reduce the nutrient emissions. Investigations of different systems all over the world show that nearly every system responses differently to eutrophication. Especially physical boundary conditions play an important role in the manifestation of the ultimate effect of local eutrophication. Apart from the physical boundary conditions also the transformation and retention of nutrients in estuarine and coastal systems contribute to system specific responses. Depending on all these different conditions, site specific responses with even site specific problems may occur. The challenge of this millennium is to really reach a balance between nature and mankind including its population size. A beneficial step in this discussion is assessing criteria to reduce eutrophication back to a level acceptable to both humans and nature.

Introduction

Eutrophication is a process that contains several aspects when considering the processes responsible for it. There are coastal areas that due to local natural characteristics show eutrophication phenomena as increased mineralization of organic matter and decreased oxygen concentrations (Fig. 1). Characteristics responsible for it are the geomorphology of the systems (percentage of subtidal and intertidal areas), shape of the tidal curve (rate of change in current velocities during flood and ebb), mean depth of entire system, its channels and tidal flats (Van Straaten & Kuenen, 1958; Postma, 1967). Apart from natural eutrophication also human induced eutrophication occurs due to nutrient loadings from point sources (outfall discharges of industrial plants and sewage treatment works) and of human-influenced ‘diffuse sources’ (run-off from any but specifically an agricultural catchment). Point source discharges are usually relatively easy controllable while diffuse and atmospheric sources are more difficult to control. The last

1. Natural conditions
    - tide associated accumulation of organic matter
    - sedimentation areas
2. Anthropogenic influence

    Input from :
    - point sources
    - diffuse sources

Figure 1. Causes of eutrophication.

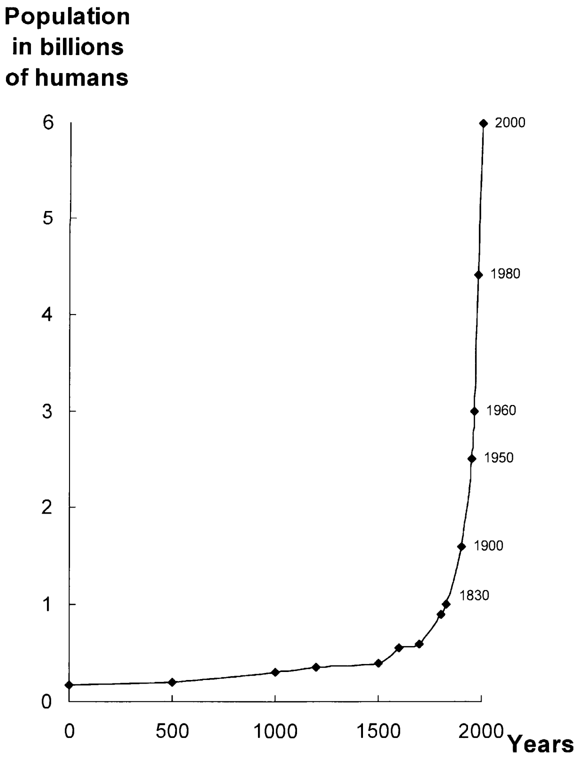

Figure 2. Increase in the world human population since era.

ones require a change in agricultural and technical practice. It is clear that the extend of the nutrient loads is directly dependent on human activities which in turn is dependent on the development of the world's human population. Human induced eutrophication is consequently in a way related to the increase of the human population. This increase is today dramatic compared to the available historic information (Fig. 2). To date, the human population has increased over 35-fold to 6 milliard individuals. The growth rate of the population has increased from 0.0002 in the period between 0 and 1500 AD to 0.071 in the period 1950 to 2000 AD, a factor of over 350-fold. According to the present data, we will have reached the level of 8 milliard people within 30 years time.

There is in general a good qualitative understanding of the processes that contribute to eutrophication. The quantitative influence on the ecological processes and the changes in community structure are, however, still not well understood.

Nutrient inputs are indispensable for the functioning of aquatic systems. Occurrence of eutrophication effects indicate when the system cannot cope with the available internal and or external nutrient inputs. Areas with low hydrodynamic energy conditions or with other suspended matter accumulation stimulating mechanisms (lagoons, estuaries and enclosed seas) can in a relative way be considered as naturally organically enriched. Consequently, little additional material is required to make them eutrophic. In contrast, there are also naturally oligotrophic areas which drain poor upland areas and receive little organic matter.

The inputs of nutrients may lead to both observable and less obvious undesired phenomena which include turbid and foul smelling (mephitic) waters, foam on the beach, oxygen depletion accompanied by a mass mortality of animals and accompanying H_2S smell, proliferation of opportunistic macrophyte algae such as representatives of the genera *Enteromorpha* and *Ulva*, the development of noxious algal blooms resulting in DSP, ASP or PSP (respectively, diarrhoeic, amnesic or paralytic shellfish poisoning) or possibly even the occurrence of the recently discovered 'phantom' dinoflagellate (*Pfiesteria piscicida*) along the east coast of the U.S. (Burkholder et al., 1992).

Since era low lying areas near the river and the coasts of the sea have been experienced to be very attractive for transport purposes and thus also for human settlement. This has resulted in a situation where these areas have been developed in densely populated and highly urbanized areas all over the world. Today, with an increasing quality of infrastructure, the human populations in these low lying areas will dramatically continue to increase, e.g. within 10 years, over 75% of the US population will live within 75 km of the coast. In developing countries, there is a continuing migration from rural, inland areas to urban, coastal areas and these trends are exacerbated by the exponential rise in populations (cf. Fig. 2) which in turn has led to large environmental problems such as eutrophication. In addition to the above, increased and more efficient agriculture has included more efficient drainage and thus the increased rate of run-off of organic matter and nutrients which contribute to high chlorophyll and productivity in estuaries (e.g. Cloern, 1991) and estuarine areas like the Dutch Wadden Sea (de Jonge, 1990, 1997; de Jonge et al., 1996). The increased production of organic waste by agriculture and increasing human population includes excess nutrients from domestic (human-induced eutrophication), intensive arable and pastoral farming and, since the late 1960s, the widespread use of artificial fertilisers in agriculture and domestic and industrial detergents. Different land uses significantly influence the release of sediment, nitrogen and phosphorous from a catchment as was shown for the Chesapeake Bay area (Fig. 3).

The consequence of the characteristics of many coastal areas is that about 28% of the total global primary production takes place here, while the surface area of these systems covers only 8% of the earths surface (Holligan & de Boois, 1993). This means that the effects of eutrophication are most manifest in the coastal zone, including estuaries.

In this introductory paper to the ECSA symposium 2000 "Managing Eutrophication of Estuaries and Near shore Waters: a Challenge for the New Millennium", we focus on the causes and mechanisms of eutrophication as well as the consequences. We give examples varying from eutrophication caused by freshwater inflow to that caused mainly by atmospheric inputs and nutrient import from the sea instead of land and atmosphere. It will be shown that increased nutrient enrichment may lead to organic enrichment and consequently to dystrophication (the modification of bacterial activity) leading ultimately to hypoxia and anoxia.

Historical information

Nutrient enrichment of the aquatic environment has always occurred but the general problem of eutrophication' has been recognised only since the 1800s. Waste deposited in the streams and rivers either stimulated in a positive way the productivity of the aquatic systems or negatively influenced systems by, e.g., structural changes in community structure (species composition) and the occurrence of hypoxia (oxygen depletion) and anoxia (total lack of oxygen). Larger bodies of water such as estuaries, the coast or enclosed seas have a greater assimilative capacity and thus require greater amounts of organic matter to create deleterious effects. For example, C, N and P enrichment has been demonstrated in Chesapeake Bay (U.S.) (Cooper & Bruch, 1991) where enrichment of sediments could be traced back to colonial times. This increase in the carbon content of the sediments could be related to eutrophication since ca. 1800 when the local population started to increase. Andrén (1999) recently reported changes in the diatom flora of the Oder estuary (Germany/ Poland; Europe) since the early 1900s and that this could also be related to increased eutrophication of the heavily populated lower reaches of the river system and its estuary.

In Narragansett Bay (U.S.A.), pre-development nutrient inputs and productivity assessments (Table 1) (Nixon, 1997) suggest that the system presumably was nitrogen limited and that the total dissolved inorganic nitrogen (DIN) input to the system has increased 5-fold and that of dissolved inorganic phosphorous (DIP) 2-fold. During the pre-development period, most of the nutrients (77–93% of DIN input and nearly 100% of the DIP input) probably entered the bay from the ocean by the estuarine water circulation rather than from the drainage basin. This feature was also postulated for European estuaries and the Dutch Wadden Sea some 40 years ago (Postma, 1954). All of these reported values are the best possible historical values but are still approximations. The 'pre-development' primary production for U.S. coastal waters (i.e. before ca. 1800) was obtained by a functional regression of field and experimental data and estimated to have approximated circa 130 g C m^{-2} a^{-1}.

For the rivers Rhine and Ems, the shallow coastal Dutch Wadden Sea and the estuary of the river Ems (Germany, The Netherlands; western Europe), the 'natural background' inputs and concentrations have been calculated (Table 1) given the present main coastline and the present reduced volume and residence time of the connected freshwater systems (van Raaphorst & de Jonge, 2000). These concentrations very well approximate the few reliable values that were published for the early 1930s (see data in Laane et al., 1993), viz. before the large-scale introduction of artificial fertilisers and detergents and the widespread connection of sewage systems to open waters. The data in Table 1 show that there has been a very large increase in nutrient concentrations in fresh water and consequently in its inputs and concentrations in the coastal areas. In comparison to the 'natural background concentrations', the input of total nitrogen increased 12-fold for the river Ems and its estuary, 4-fold for the Baltic (Scandinavia) and 10-fold for the river Rhine. Compared to the 'pre-industrial period'

Table 1. Recent and 'prehistoric' loadings of some systems by nitrogen and phosphorus and the resultant annual primary production of these systems

System (period/ year)	N influx (mmol m^{-2} a^{-1})	P influx (mmol m^{-2} a^{-1})	Mean annual nitrogen concentration in system (μmol l^{-1})	Mean annual phosphorus concentration in system (μmol l^{-1})	Annual primary production (g C m^{-2} a^{-1})
River Rhine & river Ems					
'background' situation			45 ± 25 (tN)	1.8 ± 0.8 (tP)	
early 1980s					
early 1990s					
English Channel					
'background situation			5.5 ± 0.5 (winter NO_3)	0.45 ± 0.05 (winter DIP)	
early 1930s					
early 1980s					
early 1990s					
North Sea					
'background' situation			9.1 ± 3.1 (NO_3) (near coast)	0.57 ± 0.13 (DIP) (near coast)	
early 1930s					
early 1980s					
early 1990s					
Dutch western Wadden Sea					
'background' situation			13 ± 6 (tN) ca. 4 (DIN)	0.8 ± 0.3 (tP) ca. 0.3 (DIP)	<50
early 1980s					
early 1990s					
Ems estuary					
'background' situation	(rivers) 315 (tN)	(rivers) 16 (tP)	(for salinity gradient) 10–45 (tN)	(for salinity gradient) 0.7–1.8 (tP)	
early 1980s	3850 (tN)	90 (tP)			
early 1990s	3850 (tN)	50 (tP)			
The Baltic Sea					
'background' situation (ca. 1900)	57 (tN) (rivers+AD+fix)	0.8 (tP) (rivers + AD)			80–105
early 1980s	230 (tN) (rivers+AD+fix)	6.7 (tP) (rivers + AD)			135
Narragansett Bay					
'prehistoric' situation	18 – 76 (DIN) (rivers + AD) 270–330 (DIN) (+ sea input)	~ 1 (DIP) (rivers + AD) 61 (DIP) (+ sea input)			130
'recent' (1990s)	1445 (DIN) (rivers + AD) 1725 (DIN) (+ sea input)	73 (DIP) (rivers + AD) 140 (DIP) (+ sea input)			290
Long Island Sound					
1952	-	-			ca. 200
early 1980s	1040 (tN) (rivers + AD)	70 (tP) (rivers + AD)			300
Chesapeake Bay					
mid 1980s	290–2140 (DIN) 1430 (tN)	30 (DIP) 40 (tP)			400–600

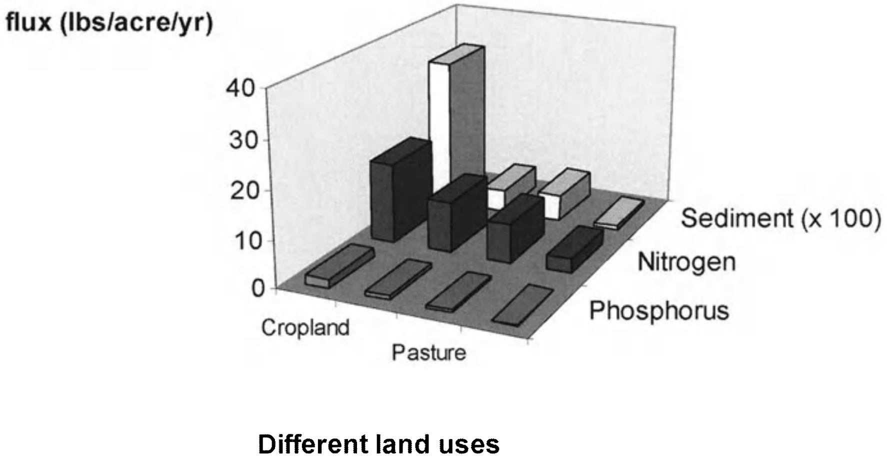

Figure 3. Release of sediment, nitrogen and phosphorus from catchment to Chesapeake Bay (modified after data from Magnien et al., 1995).

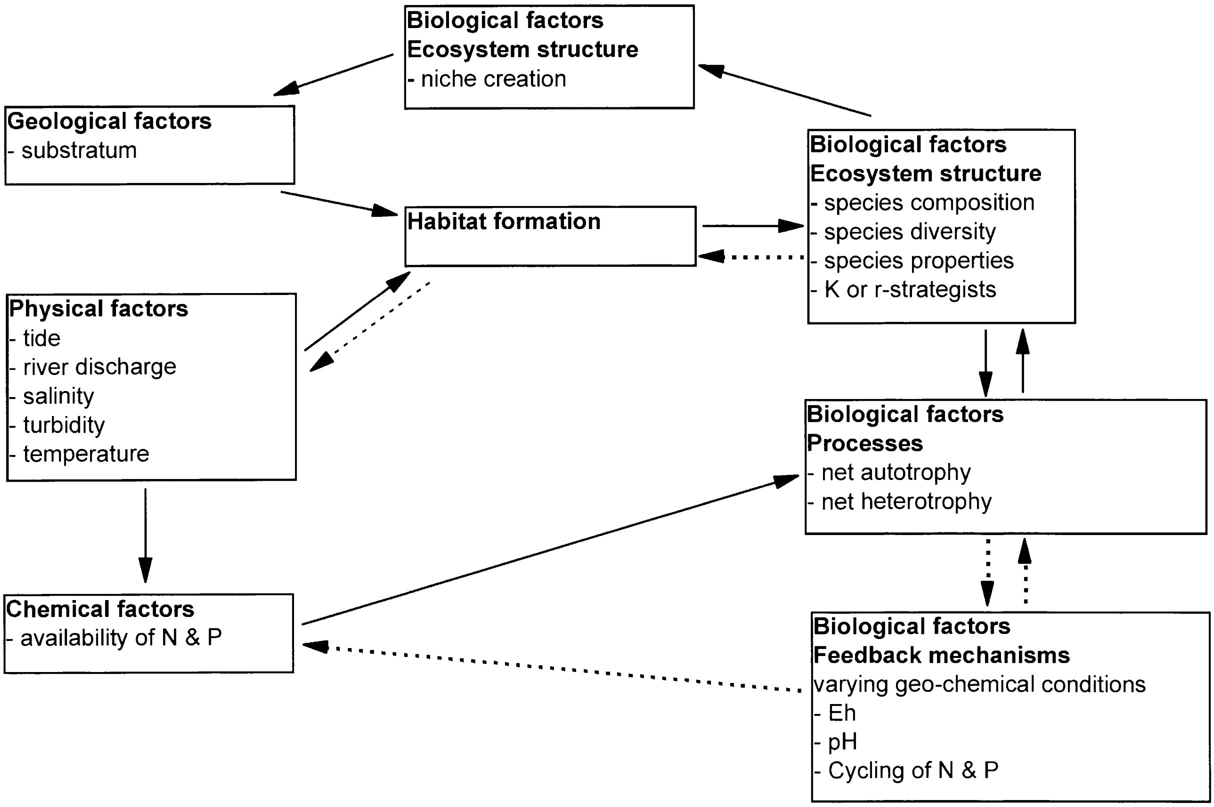

Figure 4. Operating forcing variables in the development of estuarine and marine biological communities.

the loads of total dissolved nitrogen (DIN) increased over 19-fold for Narragansett Bay (USA).

For total phosphorous (Table 1), the increase was over 8-fold for the Baltic, 6-fold for the river Ems and its estuary and 12-fold for the load of the river Rhine. Compared to the ‘pre-industrial period’ the phosphate (DIP) loads possibly increased over 50-fold to Narragansett Bay. The examples given above cover the development period of the now developed countries. Consequently, the future industrialisation and urbanisation of presently less-developed countries is likely to give a future similar trend.

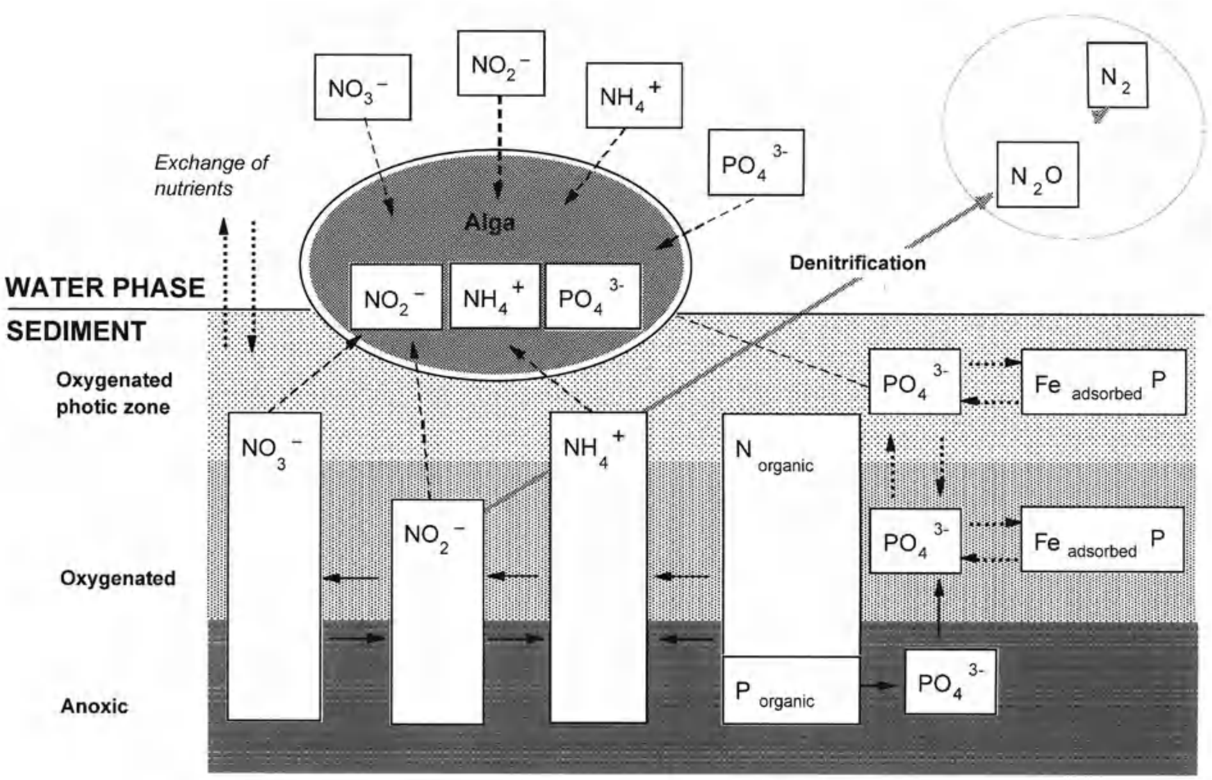

Figure 5. Schematic representation of conversion processes of nitrogen and phosphorus. Dashed lines represent assimilation, full lines represent biotic conversion (mainly microbial) and dotted lines represent geochemical equilibria. (Modified after Wiltshire, 1992 and van Beusekom & de Jonge, 1998 and references therein).

The differences in loads over the different systems are an important function of the land use (today as well as in the past). The effect of land use on nutrient and sediment release (Fig. 3) shows the absolute as well as the relative differences in these fluxes for the U.S.A. which also will be relevant for other countries with a comparable land use and application of modern equipment. The changed loads per system over time reflect the historical development in human activities and thus the changes in land use while for recent years the effect of P-reduction measures (cf. values for 1980s and 1990s for rivers Rhine and Ems in Table 1) are also notable.

Structuring elements and processes

In addition to the inputs, the mechanisms and processes which will influence the fate and effects of excess nutrient inputs will be considered. In coastal systems, eutrophication will influence not only the nutrient-related processes of the system but will also affect structural elements of the ecosystem.

Structuring elements

The physical and chemical characteristics mainly create the basic habitat conditions and niches of the marine system to be colonised with organisms. These conditions also determine colonisation rate which is dependent on the organisms' tolerances to environmental variables. Following this, the biological processes such as primary production and decomposition can modify the physico-chemical conditions while the biological inter-relationships such as reproduction, recruitment and predator–prey cycles can modify the community structure which in turn can have further consequences for the modification of the physical and chemical system in the water phase and sediment (Fig. 4). In the case of eutrophication, these processes can be summarised by the nutrient levels producing conditions for high primary production and consequent degradation possibly modifying the system through dystrophication.

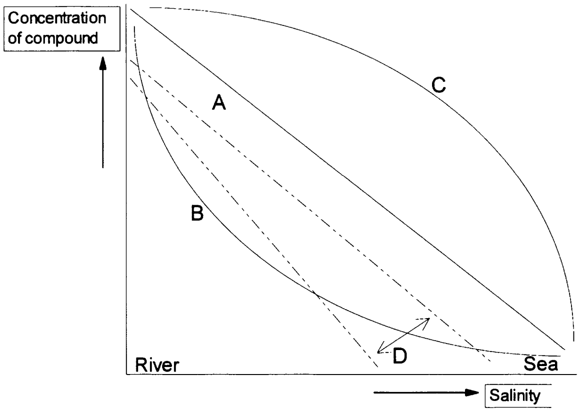

Figure 6. Typical concentration gradients between the non-tidal fresh river water and the sea. Dilution lines. (A) conservative dilution of substance within the area, (B) condition with removal of substance between the end members, (C) situation with addition of substance, (D) Illustrating different levels and slopes due changes in input and transformation rate in the system under consideration (Estuarine gradients after Officer, 1979; tidal river spikes after van Beusekom & de Jonge, 1994).

The autochthonous inputs of organic matter to the estuarine and coastal systems include reed beds, salt marshes, phytoplankton, microphytobenthos (benthic micro algae) and macro algae, whereas the allochthonous inputs are from riverine, atmospheric and marine sources, terriginous-blown material and anthropogenic inputs such as sewage. Each of these sources supplies organic matter to the system and many of the former will retain nutrients for a short period before being degraded and releasing the nutrients through the microbial loop.

Processes

As a transition area, estuaries play a filtering role for both dissolved and particulate matter, including the compounds associated to it. These estuarine areas can be considered as a reactor vessel with input, transformation, sink and output functions related to organic carbon and nutrients. Part of the transformation processes is illustrated in Figure 5 (Wiltshire, 1992; van Beusekom & de Jonge, 1998 and references therein). From the figure presented it is clear that estuaries may operate as sources and as sinks of organic matter and nutrients.

Import

The import from especially the sea is difficult to assess. This is due to the fact that the net transport of the several compounds is only a very small fraction of the total amounts moving in and out with the flood and the ebb tide. In the Ems estuary, the annual import of organic carbon was for example estimated to be only 4.5% of the annual longitudinal flux through the tidal inlet (de Jonge, 1995). Similarly, the river import to an estuary is poorly known and difficult to assess especially as in the freshwater tidal zone the mud from either marine or terrestrial origin alternately is in contact with either fresh or brackish water over a single tidal cycle. Due to the differing ionic composition of these water bodies at the freshwater–brackish water interface, sudden and extreme variations in physico-chemical conditions result in rapidly changing adsorption–desorption equilibria and consequently spiking of concentrations of several elements as dissolved Fe, dissolved Al, DIP, Eh, pH, dissolved organic carbon (Fig. 6). A possible solution for the problem of calculating the river input was presented by Officer (1979) and was based on calculating the river inputs based on estuary reaches with apparent conservative nutrient behaviour.

Transformation

Within the estuarine system, the shape of gradients in nutrients and suspended matter, and also the slopes and levels of it all vary over different time scales. General patterns are, however, observable. Important large-scale transformations take place in which biological (uptake of nutrients by primary producers, transformation by grazers and degradation by detrivores) as well as geo-chemical processes (adsorption-desorption, precipitation and dissolution) and burial of substances play a role. This produces a model of conservative behaviour and the illustrative deviation by nutrient measurements from that behaviour. In the absence of any operating process, except dilution, any concentration gradient of a substance with relatively high concentrations at the freshwater end-member and relatively low values in the sea would be a straight line (curve type 'A' in Fig. 6). When, however, significant removal of the substance occurs, due to e.g. transformations or deposition, the curve type 'B' will appear while during significant addition of substances, by point sources or local transformation processes, curve type 'C' will emerge. However, the addition or withdrawal of substances over the entire system will also change the slope of the gradient or even the level of the entire gradient ('D' type in Fig. 6).

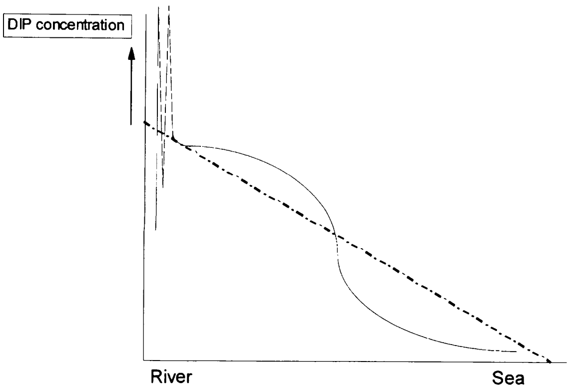

Figure 7. Typical concentration gradient of dissolved inorganic phosphate (DIP) in a turbid mesotidal estuary with a mean flushing time of ca. 30 days.

The concentration gradients of total dissolved nitrogen (DIN) and reactive silicate are often of the 'A' type (Fig. 6), while those of reactive dissolved phosphate (DIP) often experience distributions of the 'B' type as well as the 'C' type or even mixtures (Fig. 7). This is due to transformation processes (a combination of biological, geochemical and physical processes operating) and the strong reactivity of phosphate in aquatic systems. This also indicates that the behaviour of silicate, nitrogen and phosphorous differ in aquatic coastal systems. The cycle of nitrogen is almost completely determined by biological processes, that of silicate by a combination of physical processes (e.g. the physically determined dissolution of diatom frustules) and geo-chemical conditions (adsorption–desorption processes) while the phosphorous cycle is strongly determined by biological processes in combination with geo-chemical processes (adsorption–desorption processes and apatite formation). The basic pathways of the elements N, P and Si is presented in the simplified cycles of Figure 8.

The governing processes with the dominant effect depends on physical factors such as the input of nutrients, the turbidity of the system, the residence time (see below) and occurrence of stratification (Fig. 9). Systems with low turbidity are more sensitive to increased nutrient concentrations than highly turbid systems as productivity is directly governed by light extinction coefficient. With increasing natural turbidity of coastal systems, this phenomenon becomes proportionally less important. For example, in most of the North Sea estuaries which have a notable turbidity maximum zone in their upper reaches, it does not play any role in the upper reaches but does in the lower reaches cf. de Jonge, 2000). It is, however, extremely important for fjordic systems (Norway, Scotland), the Baltic Sea and very sheltered estuarine systems as found in the U.S.A. Similarly, stratification causes different physico-chemical conditions which may result in hypoxia or anoxia and corresponding processes such as hypoxia-stimulated denitrification and, due to low redox conditions, high concentrations of DIP (Fig. 5). The produced compounds may be transported to the surface layers where they will stimulate process rates.

Retention

There are two main mechanisms that stimulate retention of nutrients: denitrification (output of nitrogen to atmosphere) and burial (Fig. 9). Only the latter represents the physical retention of substances. The residence time or the flushing time of fresh water determines the degree of retention of fresh water and implicitly also determines how much of the available nutrients will be removed from the system by incorporation and storage as plant tissues, to the atmosphere (denitrification) or to the bottom sediments (by burial). All systems show, dependent on flushing time and geomorphologic characteristics, a certain removal or disappearance of nutrient compounds.

Output

Denitrification may significantly contribute to the removal of nitrogen under favourable conditions. There are indications that denitrification and nitrification are stimulated by the existence of oxygenated and hypoxic water masses (and sediment pockets) close to each other. The relative nitrogen removal by denitrification ranges between 20 and 50% of the annual input (Fig. 10 after Seitzinger, 1988 modified and supplied with recent data). This suggest a wide variation in bacterial processes enhancing the transformation of nitrogen. The presented values are, however, based on different approximations and measuring methods so that a good and close comparison of the data presented so far is difficult. Newer methods lead to higher figures.

Nutrient mass balances show remarkable differences between systems. In Table 2, values are presented for three systems which largely differ in size (Ems estuary, Chesapeake Bay and Baltic Sea). These systems are situated in western countries with relatively high population density. Important differences emerge

Table 2. A phosphorus and nitrogen budget for the Ems estuary for 1992/93 (van Beusekom & de Jonge, 1998) and for the Chesapeake Bay (Boynton et al., 1995) and the Baltic Sea (original data from Larsson et al., 1985, cf. also de Jonge et al., 1995)

Ems estuary	Phosphorus		Nitrogen	
(463 × 10^6 m$^{2)}$	(%)		(%)	
τ_f: 0.11 yr				
Mean K_z: 8.2–0.8	Input	Output	Input	Output
Rivers	71.4		92.3	
Point sources	~ 0			
Atmosphere	1.4	0.4	1.5	20.2
N-fixation				
Sea (dissolved)		59.3		77.1
Sea (particulate)	27.3	6.2		
Sediment (burial)		40.3		2.7
Water: accumulation				
Fisheries harvest	~ 0			
TOTAL	44.7 (100%)	44.7 (100%)	2362 (100%)	2278 (100%)
(× 10^6 mol yr^{-1})	96 mmol m^{-2} a^{-1}		5100 mmol m^{-2} a^{-1}	
Chesapeake Bay	Phosphorus		Nitrogen	
(11 478 × 10^6 m^2)				
τ_f: 0.5 yr				
Mean K_z: 3.0–0.75	Input	Output	Input	Output
Rivers	43.7		59.7	
Point sources	26.4		28.3	
Atmosphere	4.8		12.0	26.0
N-fixation				
Sea (dissolved)		0		30.3
Sea (particulate)	25.1			
Sediment (burial)		96.3		34.8
Water: accumulation				
Fisheries harvest		3.7		8.9
TOTAL	485 (100%)	485 (100%)	10834 (100%)	10 834 (100%)
(× 10^6 mol yr^{-1})	42 mmol m^{-2} a^{-1}		944 mmol m^{-2} a^{-1}	
The Baltic Sea	Phosphorus		Nitrogen	
(373 000 × 10^6 m^2)				
τ_f: 22 yr				
Mean K_z: 'clear'	Input	Output	Input	Output
Rivers	65.5		54	
Point sources	26.5		8	
Atmosphere	7.2		27	70–75
N-fixation			11	
Sea (dissolved)		10		10
Sea (particulate)				
Sediment (burial)		70		5–10
Water: accumulation		20		10
Fisheries harvest				
TOTAL	2500 (100%)	2500 (100%)	85 000 (100%)	85 000 (100%)
(10^6 mol yr^{-1})	6.7 mmol m^{-2} a^{-1}		228 mmol m^{-2} a^{-1}	

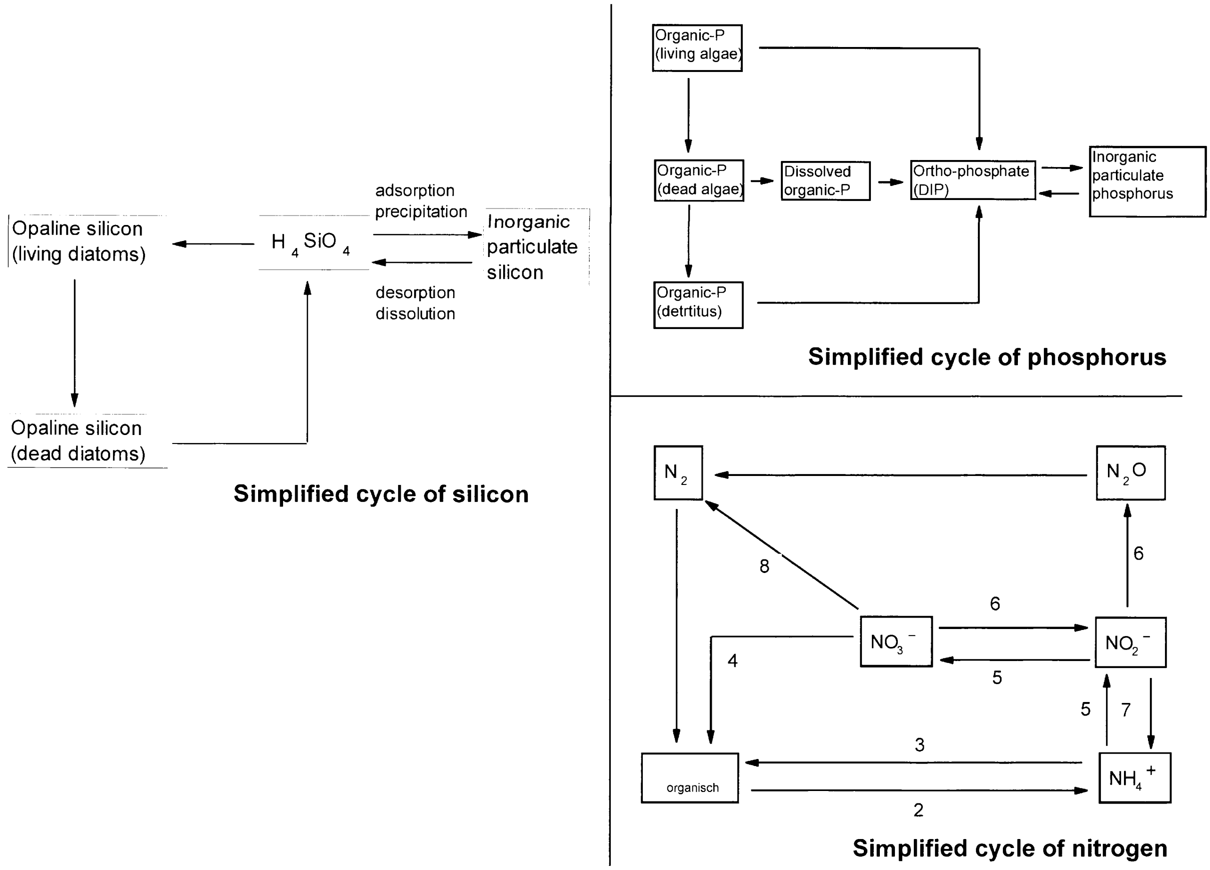

Figure 8. Cycles of N, P and Si. Explanation of the number in the N-cyclus: 1. Nitrogen fixation 2. Aerobic or anaerobic degradation or ammonification 3. Ammonium assimilation by algae 4. Nitrate assimilation by algae (at low ammonium concentrations) 5. Nitrification by chemo-autotrophic bacteria: a: $NH_4^+ + 1\ 1/2\ O_2 \rightarrow NO_2^- + 2\ H^+ + H_2O$ b: $NO_2^- + 1\ 1/2\ O_2 \rightarrow NO_3^-$ 6. Dissimilative nitrate reduction as denitrification 7. Dissimilative nitrate reduction or nitrite ammonification by a special group of bacteria (fermentative bacteria as *Clostridium* spec. and *Bacillus* spec) in strongly reduced environments as bottom sediment 8. Anaerobic ammonium oxidation (anammox) $5NH_4^+ + 3NO_3^-\ 4\ N_2 + 2\ H^+ + 9H_2O$.

when focusing on relative importance of the output of nutrients.

In the Baltic Sea and the Chesapeake Bay, both with a long flushing time, respectively, 90% and 96% of all the phosphorous influx is accumulated within the system (both sediments and water column) decreasing to only 40% for the Ems estuary due to mainly apatite formation (van Beusekom & de Jonge, 1997, 1998). For nitrogen, the differences are even greater as nearly 75% of the influx for the Baltic Sea is removed by denitrification, while in the Chesapeake Bay 35% of all the nitrogen influx is buried and for the Ems estuary most of the nitrogen influx (almost 80%) is transported to the open sea. This information shows the importance of the water column in the Baltic Sea as an accumulation compartment for nutrients.

The great differences between systems are related to the input of nutrients and to the differences in the dimensions of these systems and related factors such as the tidal range (energy) and freshwater inflow (flushing time of fresh water, residence time of fresh or seawater and turnover time of the basin water) and related important determinants as turbidity.

Despite the inherent variation, it is possible to define the most important factors in the expression of eutrophication and those that may qualitatively predict which type of system is sensitive to eutrophication (Fig. 11). The most important factors are: flushing time (f_T), the turbidity (gradient) expressed as light extinction coefficient (k_Z), and the input (external as well as internal turnover flux) and consequent concentration (gradient) of the nutrients N, P and Si. The

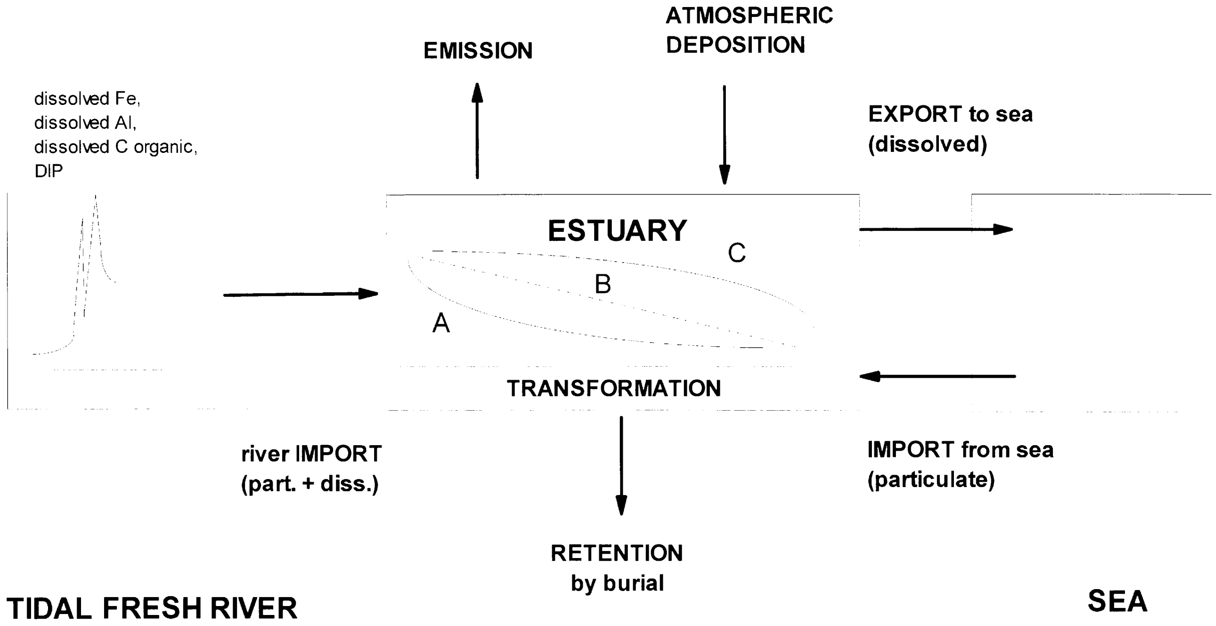

Figure 9. Diagram representing the main governing processes in an estuary.

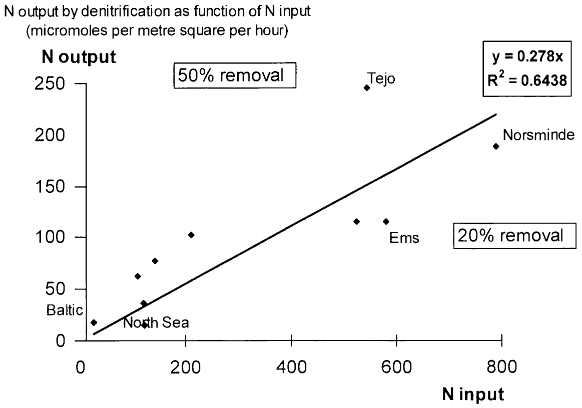

Figure 10. Scatter plot with regression line of denitrification as a function of N input of a number of systems. The systems indicated show removal between 20 and 50% of the input. Figure partly based on Nixon et al. (1996) and new data. Ems estuary (van Beusekom & de Jonge, 1998), Norsminde Fjord (Nielsen, K, L.P. Nielsen & P. Rasmussen, 1995), North Sea (Lohse et al., 1996), Wadden Sea (Kieskamp et al., 1991) and Western Scheldt (Middelburg et al., 1996).

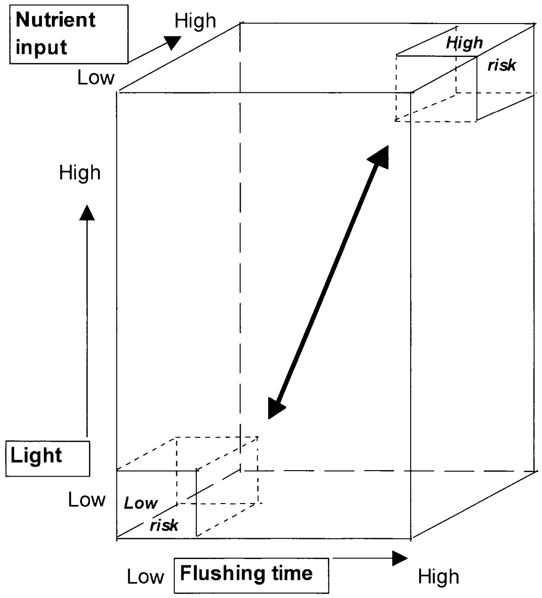

Figure 11. A 3-D classification scheme of eutrophication risk of estuaries based on flushing time, turbidity and nutrient input.

combination of mainly these three factors determines whether an estuarine system has a low or high risk of producing eutrophication symptoms.

The longer the flushing time, then the more vulnerable the system is to nutrient enrichment as the primary producers have a greater period to utilise the excess nutrients. If the flushing time is shorter, than the mean growth rate of the algae (areas with high dispersion capacity), flushing of the population will occur and thus prevent problems within the system although transport to the open sea will increase. Hence, if algal blooming does not occur within the estuary it may develop in the lowest reaches of the system or even just outside the system in the sea. A factor not included in Figure 11 is the mean depth of the system and its mixing state (stratified or not) which, in combination with (f_τ), (k_Z) and nutrient influx, may indicate the sensitivity of any system to eutrophication.

Trends and symptoms in eutrophication

Trends in eutrophication

Several trends indicate changes that have occurred in systems recently or since era. In doing this we may follow the diagram in Figure 9, thus considering input values, gradients within the estuary and output values. The symptoms following from these changed input values will be given in a separate subsection.

An example of the changes in input values are given for one of the important European rivers, the Rhine (Fig. 12) and another one for Laholm Bay in the south-western part of Sweden (Fig. 13). The river Rhine graph shows that the maximum phosphorous loads were reached in the late 1970s and early 1980s followed by a dramatic decline due to measures taken. The nitrogen loads, however, increased from the late 1950s until the late 1980s. Since then, the values dropped back to those of the 1960s.

This picture clearly shows the delay between the two components which is mainly due to the fact that phosphorous loads are better controllable than those of nitrogen because phosphorous mainly reaches the estuary from point sources while nitrogen is mainly derived from diffuse sources.

The primary effects of these long term fluctuations in loads, which were not caused by the fluctuations in river discharge, were also variations in the concentrations of these compounds in the fresh water of the river Rhine.

The biological response of the increases in the loads and concentrations were increases in the annual primary production along the coast and in the Wadden Sea, the biomass of macrozoobenthos in the western Dutch Wadden Sea, the meat content of the blue mussel and the concentrations in the western Dutch Wadden Sea (de Jonge et al., 1996).

Apart from changes in fresh water, also the concentrations of DIN and DIP in our coastal systems, the Wadden Sea included, changed over time (de Jonge & Postma, 1974; de Jonge, 1997). Even more remarkable is that the effect of the river Rhine plume was restricted to mainly the western Dutch Wadden Sea as no clear indications for eutrophication have been found for the eastern Dutch Wadden Sea (de Jonge & Essink, 1991; van Beusekom & de Jonge, this volume)

Of great concern is that also DIN and DIP (winter values) in part of the inflowing water of the North Sea, the Strait of Dover, have been increased since 1960. Whether this increase can be ascribed to human induced inputs or to natural phenomena like local up welling or the NAO (North Atlantic Oscillation) is not yet clear, but the effects are very important. Due to political and management measures, the concentrations of DIP in the inflowing fresh water has decreased substantially locally even reaching levels that are lower than the winter levels in the Strait of Dover. When applying the simple but very illustrative conservative plot of fresh water values and those of Atlantic water it becomes clear that at present the sea may be a source for phosphorous again instead of the main freshwater bodies as it must have been under pristine conditions.

The trends in loads and in concentrations of nitrogen in the coastal waters have not strongly changed yet and consequently N/P ratios have increased dramatically in Dutch coastal waters since the successful reduction measures for phosphorous. The combination of the still high nutrient concentrations and the increasing ratios have been blamed for the occurrence of nuisance blooms.

Symptoms of eutrophication

General effects

Based on the information presented so far, it may be difficult to assess the anthropogenic impact against the background of the natural variability in nutrient concentrations and related processes. This means that also the interpretation of apparent eutrophication related symptoms is sometimes very difficult.

Increase in the decomposition of organic matter may lead to hypoxia and even anoxia although these effects are strongly dependent on the turbulence of the system.

This degradation of organic material reflect a major community response, by species changes through the loss of intolerant or *k*-selected species and the increase of opportunistic or *r*-selected fauna. Loss of vascular plants is a primary effect due to which complete communities may disappear as happened in Australia and North America. Although very often the loss of the long living and slow growing species (i.e. *k*-selected species) is compensated by the occurrence of more op-

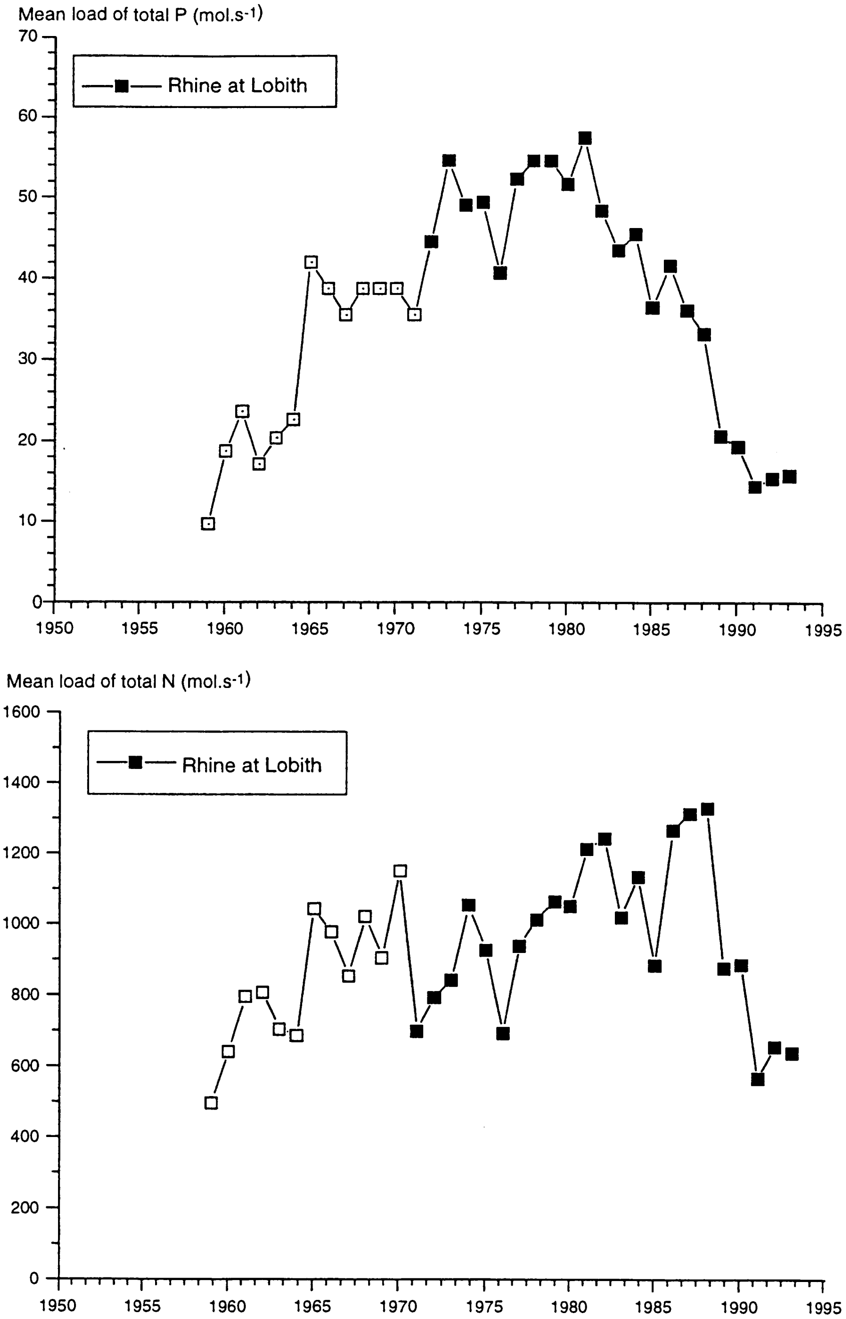

Figure 12. Development of the loads in total phosphorus and total nitrogen as measured on the border between Germany and The Netherlands (data from Rijkswaterstaat and van Raaphorst & de Jonge, 2000).

portunistic macrophytes (i.e. *r*-selected genera) such as representatives of the genera *Enteromorpha*, *Ulva*, *Chaetomorpha*, *Rhizoclonium* and *Cladophora,* this represents a deterioration of quality as these algae may form dense layers or mats associated with oxygen deficiency and high degradation rates of organic matter leading to anoxia (Raffaelli et al., 1998).

An increase in nutrient ratios may, based on differences in affinity of algae to nutrients, cause a change in the algal species composition.

The occurrence of noxious blooms of toxin-producing algae is another problem. Possibly even worse is the phenomenon of the 'phantom' algae *Burkholder et al., 1992) of which the possible re-

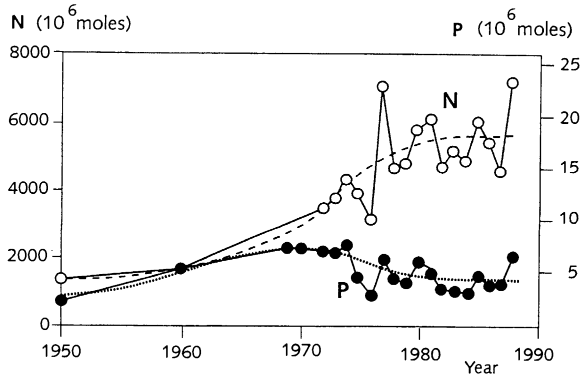

Figure 13. Nutrient loads to Laholm Bay at the south-western coast of Sweden (after Rosenberg et al., 1990) (with permission).

lation with eutrophication is yet unclear. Harmful algal blooms seem to be more common today than in the past. They can have serious effects in fish kills and neuro-toxin production (PSP, DSP, ASP) affecting animals with complex nervous systems.

The amenity value of shores is negatively influenced through foams produced by the gelatinous microalgae such as *Phaeocystis* and *Chaetoceros socialis.* The presence of the 'phantom' algae will completely destroy locally any form of recreation which will strongly affect local economy. Low turbidity systems may change from macrophyte-dominated into a phytoplankton-dominated system even with microalgal blooms affecting the integrity of the system.

In highly turbid systems where primary production is light limited, the situation usually differs from the above because benthic communities are usually poor in vascular plants like eelgrass and large brown algae. However, the effects of eutrophication from riverine sources in these systems will occur in nearby coastal area (cf. de Jonge et al., in press). Furthermore, water transport processes dictate that the effects of nutrient enrichment occur either in the lower reaches of the estuary or in the coastal area itself. Ultimately, the effect of 'habitat loss', poor water quality and a deterioration of the marine environment may negatively influence any form of human use.

Large scale effects

Symptoms of eutrophication are for instance the increase in productivity of seas as e.g. North Sea (Peeters et al., 1993) and the Wadden Sea (de Jonge et al., 1996 and references therein), the observed increase in surface algal blooms in the Dutch EEZ of the North Sea (Fig. 14), a major sudden change in plankton composition from diatoms to small flagellates (Hickel et al., 1993) and the (temporarily) large scale oxygen deficiencies in e.g. the North Sea (Zevenboom, 1993; Zevenboom et al., 1997), the Baltic, Long Island Sound and Chesepeake Bay (de Jonge et al., 1995).

Eutrophication in Florida Bay possibly produced a large scale sea grass die off (4000 ha of *Thalassia testudinum* and *Halodule wrightii* disappeared between 1987 and 1988), followed by increased phytoplankton abundance, sponge mortality and a perceived decline in fisheries (Rudnick et al., 1999). This very large change in the health of the system followed major engineering works to the Everglades area and reflected changes in the nutrient concentrations, the nutrient pool, the chlorophyll levels and turbidity. The preliminary nutrient budget for the bay assumes a large oceanic and atmospheric input of N and P to the Bay although the denitrification rates are unknown. The cause(s) of the sea grass mortality in 1987 is still unknown.

Another symptom is the dramatic increase of the permanent anoxic layer of the Baltic which rose from 18 000 to near 70 000 km^2 which is nearly the entire hypolimnion (Jonsson et al., 1990). The increase of the anoxic layer had dramatic ecological consequences. It resulted in a dramatic structural decline in population size of two important prey species (the large isopod *Saduria entomon* and the snake blenny *Lumpenus lampetraeformis*) which in turn greatly influenced the local cod populations (Elmgren, 1989). The given changes were the result of an increase in the primary production of only 30–70%. Another consequence was an increase in the density of algal mats which reduced the development of herring eggs, possibly by production of exudates. Finally, the hypoxic areas adversely affected cod egg development (Elmgren, 1989).

Sommer (1996) performed experiments with periphytic micro algae showing complex behaviour under changing ratios of Si, P and N. Relative high Si led to dominance of diatoms while relatively low Si and low N favoured the development of cyanobacteria while chlorophytes took advantage at low Si and low P. Moreover, variations in light intensity influenced the outcome.

The extensive bloom of the micro alga *Chrysochromulina* species in the late 1980s in the outer Baltic apparently developed in response to nutrient build up on the eastern North Sea and contributed to the hypoxia and eutrophic symptoms. Near the west Atlantic coast in the U.S.A. the 'phantom algae' (*Pfiesteria pis-*

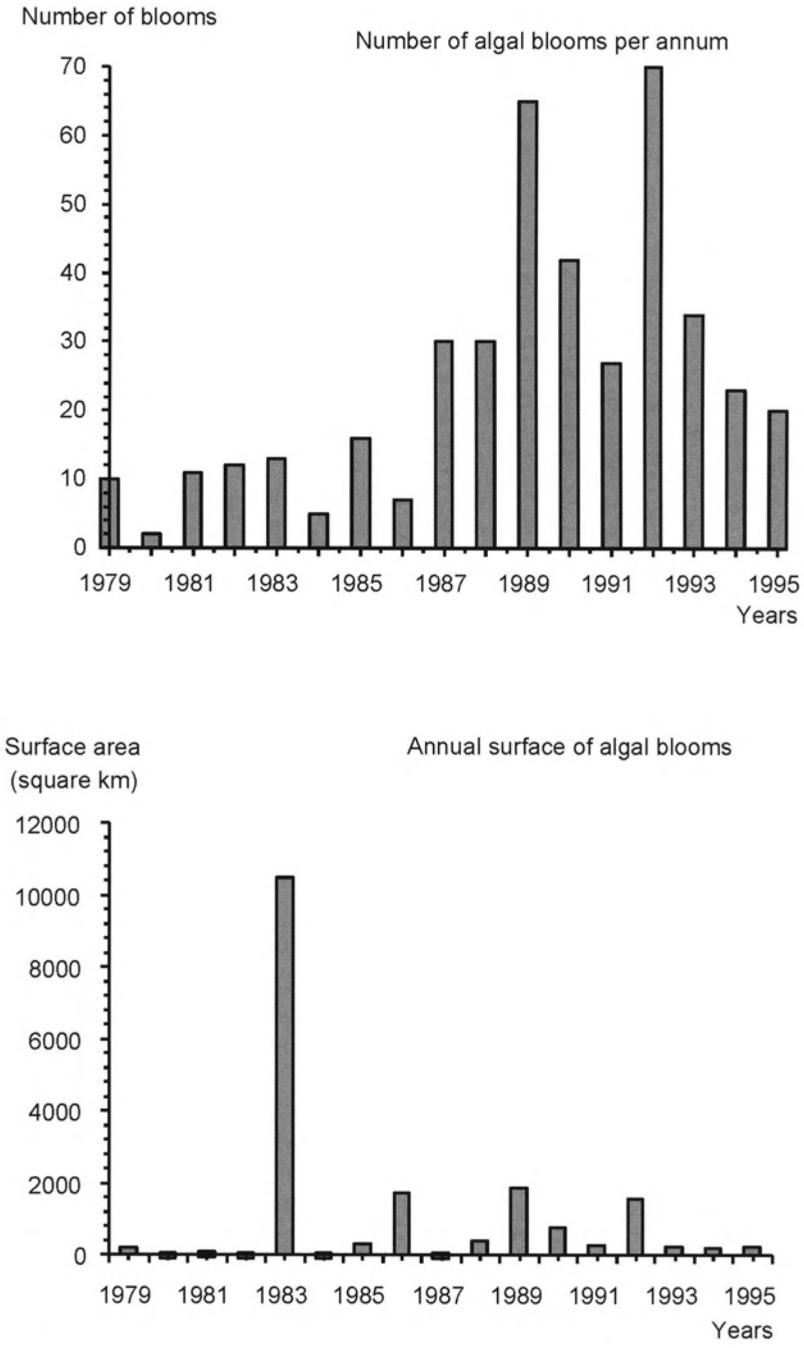

Figure 14. Surface algal blooms observed during Dutch airborne surveys over the period 1979–1995 (after Zevenboom, 1993; and Zevenboom et al., 1997) (A) frequency per annum (B) surface area in km^2 per annum.

cicida) occurred since the early 1990s (Burkholder et al., 1992).

Observed changes in primary producers generally reflected vascular plants operating as k-strategists under stress and replaced by more opportunistic algae like phytoplankton species (cf. Margalef, 1978). Although not discussed here, also synergistic effects where eutrophication effects are exacerbated by other pollutants are expected in urbanised and developed areas.

Local effects

Apart from large scale general effects as reported above, there are also strong local effects observable due to fish farming (Bonsdorff et al., 1997). These authors further stated that 35% and 55% of the total local nitrogen and phosphorous inputs are accounted for by local inputs leading to a strongly reduced N/P ratio. The local nutrient enrichment in the Baltic produced an increase in the primary production and an increase in turbidity which negatively affected the macrophyte populations as eelgrass (*Zostera marina*). It further stimulated the blooming of cyanobacteria. Loss of five benthic crustaceans have been observed while a gain of four was reported of which two were polychaetes (*Polydora redeki*, *Marenzelleria viridis*) new to the area. The macrozoobenthos community further showed a structural change from suspension feeders to deposit feeders.

The Peel-Harvey estuarine system (South Pacific West coast of Australia) receives a high nutrient loading, but has an additional phosphorous release during stratification-induced anoxia from the bottom sediments. This happens after a clear loading of the estuary and the subsequent development of dense populations of microphytobenthos which is responsible for the nutrient storage (McComb & Lakatelich, 1995). This release contributed to changes in macro algal community structure and increased turbidity due to algal blooms.

Creation of red-tides (noxious, toxic and nuisance micro algal blooms) in Tolo Harbour (Hong Kong) resulted from large urban nutrient inputs, a water residence time of 16–42 days and a low turbidity (cf. Figs 15 and 11) which led to dense phytoplankton populations. Diatoms decreased in abundance from 80–90% to 53% in 1982–85, dinoflagellates increased concurrently with red tides and chlorophyll-*a* levels also increased significantly. Oxygen depletion occurred due to nutrient loadings and the local development of phytoplankton in combination with reduced water exchange (long flushing time), features associated with a low-energy eutrophic environment (Hodgkiss & Yim, 1995).

In other systems, e.g. North Sea waters regular blooms of nuisance algae occurred of which it is assumed that these are related to high nutrient concentrations and high N/P ratios (Zevenboom et al., 1997). Among these species are *Phaeocystis* species, *Noctiluca scintillans*, *Gyrodinium aureolum*, *Dinophysis acuminata*, *Alexandrium tamarensis*, and several *Prorocentrum* species.

Effects at the conceptual level

A conceptual model of the main effects of eutrophication is presented in Figure 16 which gives the consequences of nutrient enrichment on primary production and for nutrient cycling, and shows the secondary effects of turbidity, changes in species composition and organic decomposition. Increased nutrient concentrations may lead to changes in both the species composition (lethal and sub lethal concentrations of nitrate and ammonium to e.g. sea grasses) and the primary production. The turbidity effect is particularly relevant to systems with a natural low turbidity because these often contain communities of macrophytes and are therefore sensitive to nutrient enrichment. Among these vulnerable species are macro algae and/or vascular aquatic plants (e.g. sea grasses) which have a restricted immersion tolerance and thus may be lost from the low turbidity systems in poor light conditions.

Future challenges

The variation in the percentage of denitrification of the total N input (Fig. 10), which might partly be caused by the application of different methods, suggests that it is necessary to reconsider the importance of this process.

The finding of anaerobic ammonium oxidation (Mulder et al., 1995) suggests that it might be worthwhile to further explore this and other unknown nitrogen transformation processes and routes in estuarine areas. In addition to these transformation processes and given the trend in decreasing nitrogen loads, it seems of increasing importance to study N fixation processes under estuarine conditions (e.g. Pearl, 1990; Pearl et al., 1994).

There are several recent and historical indications for the significant role of geochemical processes on extremely strong fluctuations in values of e.g. pH, Eh, DIP, dissolved Fe, Al and carbon in the freshwater tidal part of rivers which strongly influence particle composition (Morris et al., 1982, 1987) and hamper measuring of correct input data (van Beusekom & de Jonge, 1994). It is necessary to further unravel the interaction between fresh water, brackish water, river born mud and sea born mud during the tidal cycle and its impact on the above given concentrations.

The internal silicate input within estuarine systems from local, weathering of clay minerals should be de-

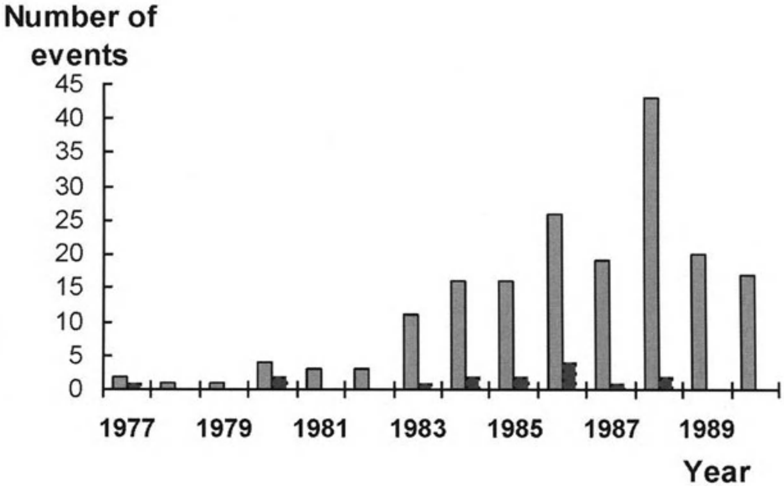

Figure 15. Red tides and associated fish kills in Tolo Harbour (after Hodgkiss & Yim, 1995; with permission).

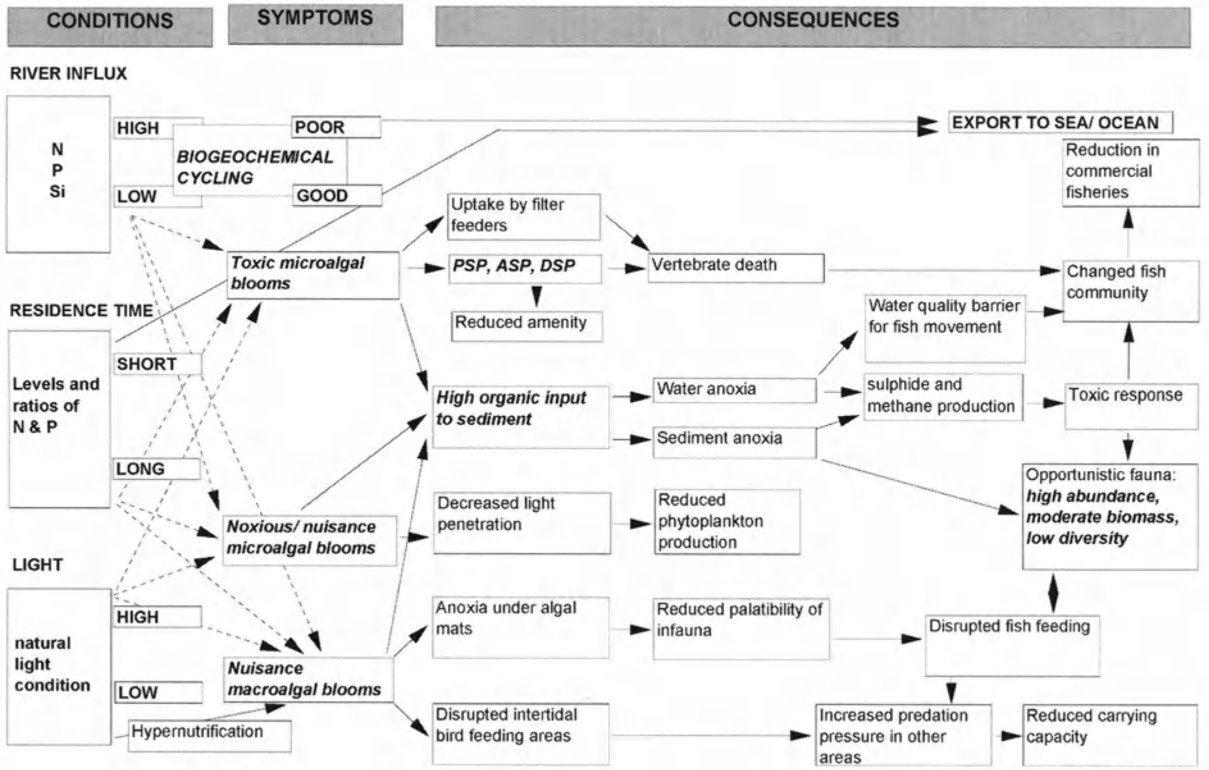

Figure 16. Overview of potential effects of nutrient enrichment in combination with turbidity conditions and residence time of water in an estuarine area.

termined. Also the role of dissolved reactive silicate in preventing the development of harmful blooms of flagellates should be investigated.

Finally, the effects of high N/P ratios should further be assessed.

Conclusions

Man has a choice of deciding how to treat natural resources, of acknowledging that organic waste (including nutrients) will continue to be produced and that there may be decisions of whether to treat it through techniques or allow natural systems the sea included to degrade it. In essence, the biological degradation occurs whether in a treatment works or in aquatic systems.

What has to be safeguarded, however, is the integrity and sustainability of the natural system undertaking that degradation. The examples here show the very great spatial and temporal changes that may occur, under the influence of our own activities and focusing on compounds and elements that are part of the system. Unlike many other pollutants, nutrients are required for the well being of the system which has a natural ability to degrade, disperse and accumulate them. Only when this natural assimilative capacity is exceeded will problems occur.

Atmospheric input, river run off of point sources and diffuse sources will further increase the concentrations of nutrients in sea water on a global scale and while past 'hot spots' have centered on point sources, which are relatively easy to control, future controls will have to concentrate on diffuse terrestrial and aerial inputs.

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Modern foraminiferal record of alternating open and restricted environmental conditions in the Santo André lagoon, SW Portugal

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Key words: benthic foraminifera, coastal lagoon, barrier, human intervention, seasonal changes, southwest Portugal

Abstract

Benthic foraminifera from eleven stations sampled seasonally were analyzed in order to examine the biological response to rapid and intense environmental changes taking place in the Santo André coastal lagoon. Foraminiferal assemblages show a very low species diversity and a high dominance of three euryhaline species throughout the year. Under closed-inlet conditions, foraminiferal assemblages exhibit low abundance of foraminiferal tests, whereas under open-inlet conditions absolute abundance greatly increases due to sea water entrance. Present-day dramatic environmental changes are thought to be responsible for deformed foraminiferal tests that are commonly found. Comparison of modern assemblages with those obtained from the Holocene sedimentary record indicates persistent restrictive environmental conditions after sandy barrier formation, circa 5000 years ago.

Introduction

The Santo André lagoon is located in the southwestern Portuguese sandy coast between Tróia and Sines (Fig. 1), an arcuate high-mesotidal littoral segment where cross-shore sediment processes dominate over longshore drift in spite of a high wave regime, due to the equilibrium shape of the nearshore.

The Santo André lagoon has an extent of 1.7–2.5 km^2, its average depth and water storage capacity varying seasonally, the maximum depth not exceeding 4 m. It consists of a central main basin which connects with a number of confined N-S elongated troughs (locally designated by 'Poços') by restricted channels (Fig. 1). The main lagoonal body is separated from the open ocean by a continuous reflective sandy barrier that may naturally breach during heavy storms allowing limited entrance of marine water. Most of the water exchange with the ocean is ensured by periodic artificial opening of an inlet on the northern area, in front of sample station 1 (Fig. 1). This procedure has been regularly undertaken since at least the 17th century in order to prevent eutrofication and to drain the tributary alluvial plains, that have been reclaimed for grazing. Nowadays the inlet is opened every year, generally in March/April, during spring tides allowing partial or complete renewal of the lagoonal water during a short period of time (typically a few weeks). The inlet silts up naturally essentially due to cross-shore sand transport healing up the barrier.

The physico-chemical parameters of the lagoonal water mass are essentially controlled by the alternating open/closed condition of the inlet. In fact, during autumn and winter the inlet is closed and the lagoon only collects fresh water from the watershed, increasing its depth and generating a stratified water mass (Freitas et al., 1998). During spring and summer, after the breaching of the barrier, the water depth decreases and the water mass is renewed and homogenized, particularly in the central basin. In this way, under summer conditions depth is lower and salinity is higher, whereas under winter conditions depth is higher and salinity decreases. In the same way, temperature and dissolved oxygen of the bottom water are higher in summer than in winter when dissolved oxygen reaches very low values (Fig. 2). This pattern

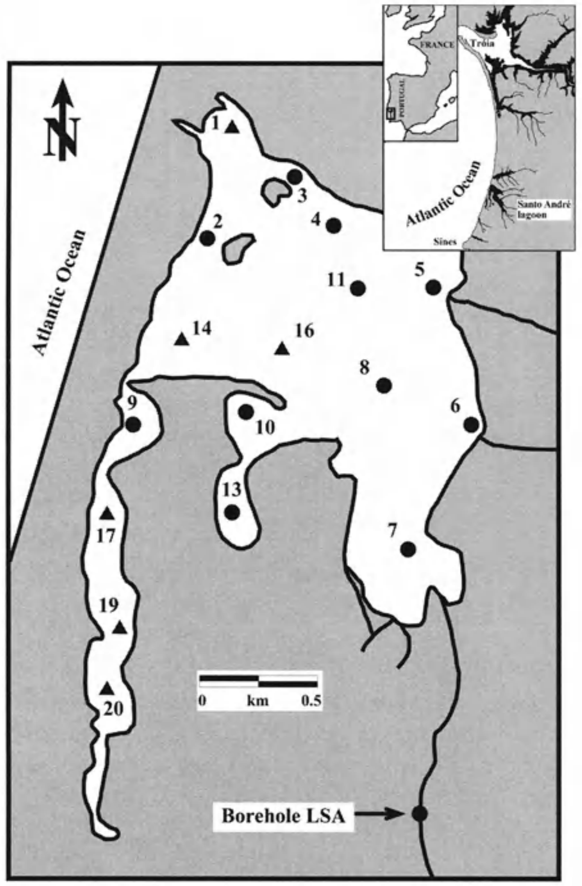

Figure 1. Geographic location of the Santo André lagoon in SW Portugal, showing the position of the surface samples and borehole. Triangles represent preliminary samples. Dots represent seasonal samples.

may be occasionally disturbed by ephemeral inputs of marine water during winter storms when overwash of the barrier occurs.

The alternation between open and closed conditions also represented a major control of the natural environmental evolution of this coastal area during the Late Holocene (Cearreta et al., in press).

The watershed involving the lagoon extends over 145 km^2; its western half is defined on a coastal plain where Cenozoic deposits outcrop and further east it includes the Flysch Palaeozoic rocks of the Serra de Grândola hills.

The main objective of this study is to examine the biological response of benthic foraminifera (hard shelled protozoa with an exclusively marine habitat: brackish, normal marine and hypersaline) to the seasonal dramatic environmental changes that take place nowadays in this coastal lagoon. An additional objective is to compare the modern foraminiferal assemblages with those found in the Holocene sedimentary record of the lagoon in order to determine the role played by the sandy barrier in the environmental evolution of the lagoon through time.

Materials and methods

Twenty sampling stations were initially selected for a general characterization (Fig. 1) and 11 locations of this set have been monitored and sampled seasonally (March, June, September and December 1999). Bottom sediments were taken with a plastic cylindrical 30 mm diameter sampler operated from a boat. Once aboard, 15 cm^3 subsamples from the surface sediment layer (0–1 cm) were collected.

Bottom sediments are sandy in the northern and eastern areas of the lagoon and muddy in the central and southern areas. They also exhibit higher organic matter and calcium carbonate contents in the latter areas due to their distal location in relation to the inlet and more restricted environmental conditions (Fig. 3). Sediments show a laminated structure probably reflecting seasonal changes of environmental controls.

In the laboratory, samples were wet sieved through a 63 μm mesh and dried. When possible around 300 individuals were picked, mounted, and identified from each sample. However, some samples were barren or did not contain enough foraminifera. Samples were originally collected for sedimentological and geochemical purposes. They were not adequately preserved in ethanol and no staining method was then possible to use in order to differentiate living from dead individuals. Consequently, these are total (living plus dead) assemblages and have been considered to be representative of the seasonal foraminiferal content due to the low energy conditions that characterize this lagoonal environment, where post-mortem transport of foraminiferal tests is presumed to be negligible. Altogether, around 8000 modern foraminifera were studied under a stereoscopic binocular microscope. All foraminiferal species identified in the surface samples are listed in Appendix 1.

In addition, the 26 m long borehole LSA (UTM – 29SNC18601511) was retrieved from the alluvial plain of Azinhal using a rotary drill in June 1998 (Fig. 1). It started 2.74 m above mean sea level and ended in Miocene basement at −21.33 m, sampling the whole post Late-Glacial infill. A description of this core, as well as sample preparation for foraminiferal analysis

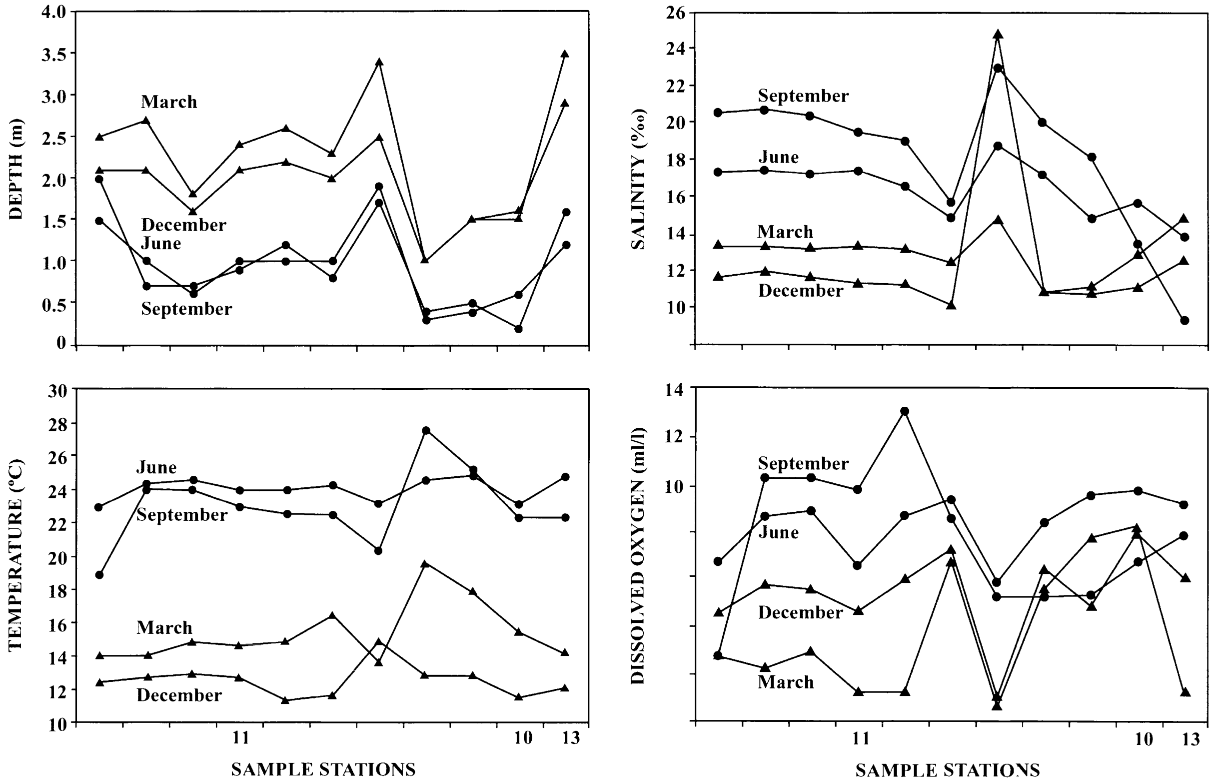

Figure 2. Physico-chemical parameters at the bottom of the lagoonal water mass in the Santo André lagoon.

and detailed results are contained in Cearreta et al. (in press).

Results

In general, abundances of foraminifera are higher in the northern and central areas (sample stations 2, 3, 4, 11, 8 and 7) than in the southern and eastern areas (sample stations 9, 5, 6, 10 and 13) of the lagoon, that are more isolated from the open ocean and receive direct fresh water run-off (Table 1).

Samples show higher foraminiferal abundance under summer conditions (mainly in June, but also in September) after the artificial opening of the inlet, related to better environmental constraints (higher salinity, temperature and dissolved oxygen contents), and lower foraminiferal abundance during winter (mainly in December, but also in March), in closed-inlet and worse environmental conditions (Table 1).

The number of foraminiferal species is generally very low in the Santo André lagoon. The highest species diversity is always found in the northern area (sample stations 2, 3 and 4), with an average species number of seven. The central area (sample stations 11 and 8) and the less confined southern stations (samples 7 and 9) exhibit an intermediate and fairly constant number of species, with an average value of three. Finally, the eastern area (sample stations 5 and 6) and the more restricted southern stations (samples 10 and 13) show a more variable seasonal species diversity, with higher values in summer (average three species) than in winter when samples can be barren of foraminifera (Table 1).

Modern assemblages of the Santo André lagoon are made exclusively of calcareous foraminifera (hyaline and porcellaneous), and no agglutinated foraminifera have been found in this seasonal study (Appendix 1).

All samples are highly dominated by three foraminiferal species, *Ammonia beccarii* (average relative abundance 55%), *Haynesina germanica* (average relative abundance 25%) and *Elphidium oceanensis* (average relative abundance 16%). Altogether they represent on average 96% of the foraminiferal assemblages found in the Santo André lagoon. *A. beccarii* is the most dominant species in nearly all samples at all seasons. *E. oceanensis* is second dominant in the northern area of the lagoon and in the eastern area after the opening of the inlet (June). *H. germanica* is second

Table 1. Seasonal relative abundance of the main foraminiferal species, number of individuals studied and number of species found in the surface samples. Black dots represent the presence of the species in samples where there were too few individuals to calculate relative abundance

Sample stations	2	3	4	11	8	7	9	5	6	10	13
March											
Quinqueloculina seminula	3.0	0.9	0	0	0	0	0	0	0	0	0
Ammonia beccarii	47.9	70.0	67.5	45.0	44.6	45.1	●	●	0	0	0
Elphidium oceanensis	30.6	22.7	25.2	10.2	5.2	12.7	0	0	0	0	0
Haynesina germanica	13.0	1.6	6.2	44.4	49.8	41.8	●	0	0	0	0
No. of individuals	330	307	274	313	365	299	5	2	0	0	0
No. of species	11	8	8	4	4	4	2	1	0	0	0
June											
Quinqueloculina seminula	12.8	16.9	8.5	0	0.9	0	0	7.9	1.4	0	0
Ammonia beccarii	46.3	39.4	44.7	51.7	50.7	36.5	14.6	43.2	57.7	●	●
Elphidium oceanensis	29.0	26.9	31.0	4.0	11.4	6.4	23.2	35.6	27.3	●	●
Haynesina germanica	6.7	10.6	13.6	44.2	36.6	57.0	61.8	13.1	11.4	●	●
No. of individuals	328	301	293	323	347	356	314	289	289	81	3
No. of species	9	8	9	3	5	3	4	4	5	4	3
September											
Quinqueloculina seminula	0.8	0.3	0	0	0	0	0	0	0	0	0
Ammonia beccarii	73.8	78.4	71.1	45.3	49.8	●	43.9	●	●	●	16.4
Elphidium oceanensis	16.1	14.3	19.1	16.6	13.2	●	4.2	●	●	0	5.1
Haynesina germanica	6.2	5.3	8.5	38.1	36.9	●	51.7	0	●	0	78.3
No. of individuals	241	313	339	289	309	45	307	15	74	1	328
No. of species	8	4	7	3	3	3	3	3	3	1	3
December											
Quinqueloculina seminula	0	0	0.3	0	–	0	0	0	–	0	0
Ammonia beccarii	63.5	76.8	77.1	71.2	–	●	●	●	–	●	●
Elphidium oceanensis	23.9	10.6	7.7	0.5	–	●	0	●	–	0	0
Haynesina germanica	10.4	6.6	11.8	28.3	–	●	●	●	–	0	●
No. of individuals	96	151	271	198	–	53	2	21	–	3	3
No. of species	4	6	8	3	–	3	2	3	–	1	2

dominant in the central area and in the less isolated southern stations (Table 1).

In June, after the breaching of the barrier, *Quinqueloculina seminula* becomes also a major species in the northern area of the lagoon (average relative abundance 13%) (Table 1). This species is an indicator of normal marine salinity conditions (Cearreta & Murray, 1996) and it practically disappears in other seasonal samples.

Most samples contain two or three deformed tests of the dominant species (Plate 1). In most cases, these deformed tests are twinned individuals.

Discussion

Numerous ecological studies in the European Atlantic coast have provided evidence that certain species of benthic foraminifera characterize particular environments. Intertidal and subtidal brackish lagoons are characterized by high dominances of hyaline species *A. beccarii*, *H. germanica*, and *E. oceanensis*. These species are highly euryhaline and typical of the southern European coastal areas (Murray, 1991). Low species diversity ranges are also typical. In the sandy areas under almost normal marine conditions, *Q. seminula* and other porcellaneous forms may be also present abundantly (cf. Murray, 1991 for a summary).

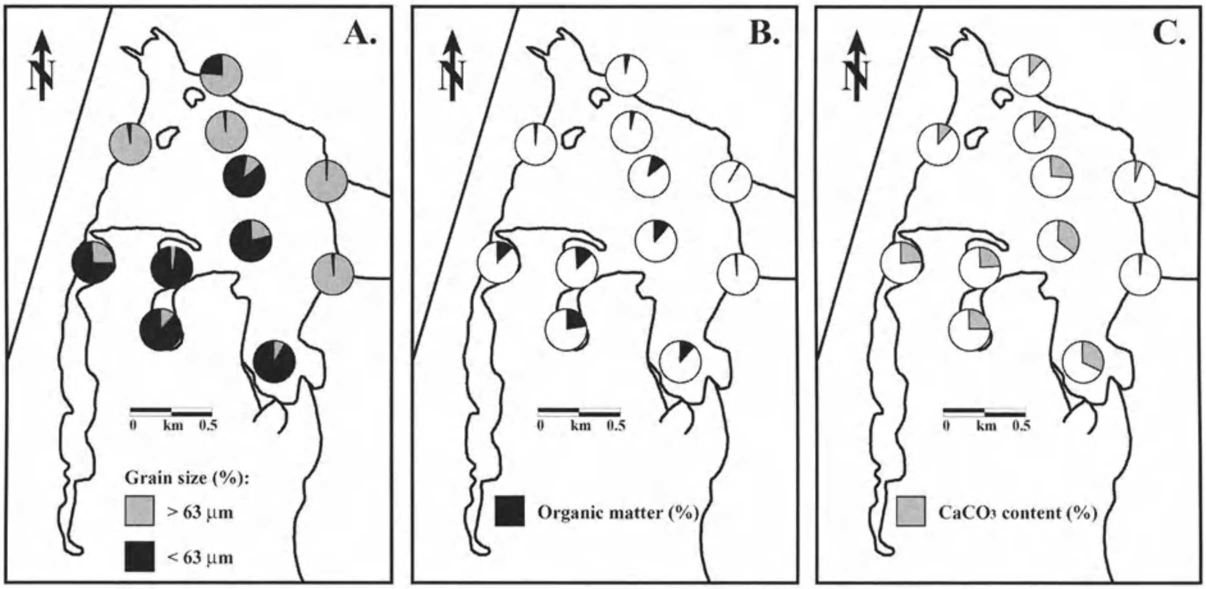

Figure 3. Texture, organic matter and calcium carbonate contents of the bottom sediments of the Santo André lagoon.

The modern foraminiferal assemblages of the Santo André lagoon show in general very low diversity throughout the year. High abundance of individuals has been found after the artificial breaching of the sand barrier whereas low abundance of foraminifera is characteristic of closed-inlet conditions. Assemblages are permanently dominated by euryhaline *A. beccarii*, *H. germanica*, and *E. oceanensis*, plus *Q. seminula* following the entrance of open ocean water into the lagoon.

Cearreta et al. (in press) studied the foraminiferal content of the LSA core. The dominant species found in this core were the same species found as dominant in the modern surface samples. On the basis of the foraminiferal contents (in terms of species abundance, diversity and dominance) the sedimentary record was divided into four major zones and seven subzones that were interpreted allowing palaeoenvironmental reconstruction. As a consequence of the sandy barrier formation (circa 5000 yrs BP), the coastal lagoon was originated and since then it has developed under restricted fresh water conditions (between 5000–3500 yrs BP and 1600–0 yrs BP) alternating with restricted, low oxygen, brackish conditions (between 3500 and 1600 yrs BP). Abundant, low-diverse, pyritized foraminiferal assemblages are characteristic of this brackish foraminiferal subzone.

Deformed foraminiferal tests (twinned individuals) are commonly found in most modern samples. However, no deformed foraminiferal tests were found in the borehole assemblages probably due to the more lasting environmental conditions in the lagoon during the Holocene compared with the modern environment where human intervention modifies seasonally the physico-chemical features of the lagoonal water mass. Twinning has been also observed in other coastal areas around the world, and has been related to rapid and intense environmental variability under natural conditions (cf. Boltovskoy et al., 1991 for a review).

Summary and conclusion

Both in the past and recent times, isolation from the open ocean and low oxygen environmental conditions have characterized the Santo André lagoon. Alternation of fresh and brackish water conditions through time due to sandy barrier development, 5000 years ago, produced a restrictive environment where extremely low oxygen conditions provoked post-mortem pyritization of foraminiferal tests. Nowadays, rapid and intense environmental changes that range from low-salinity and low-oxygen bottom water under closed-inlet conditions, to brackish and well-oxygenated bottom water under open-inlet conditions seem to be responsible of foraminiferal assemblages characterized by extremely low species diversity, high species dominances, seasonal variable abundances and common deformed tests.

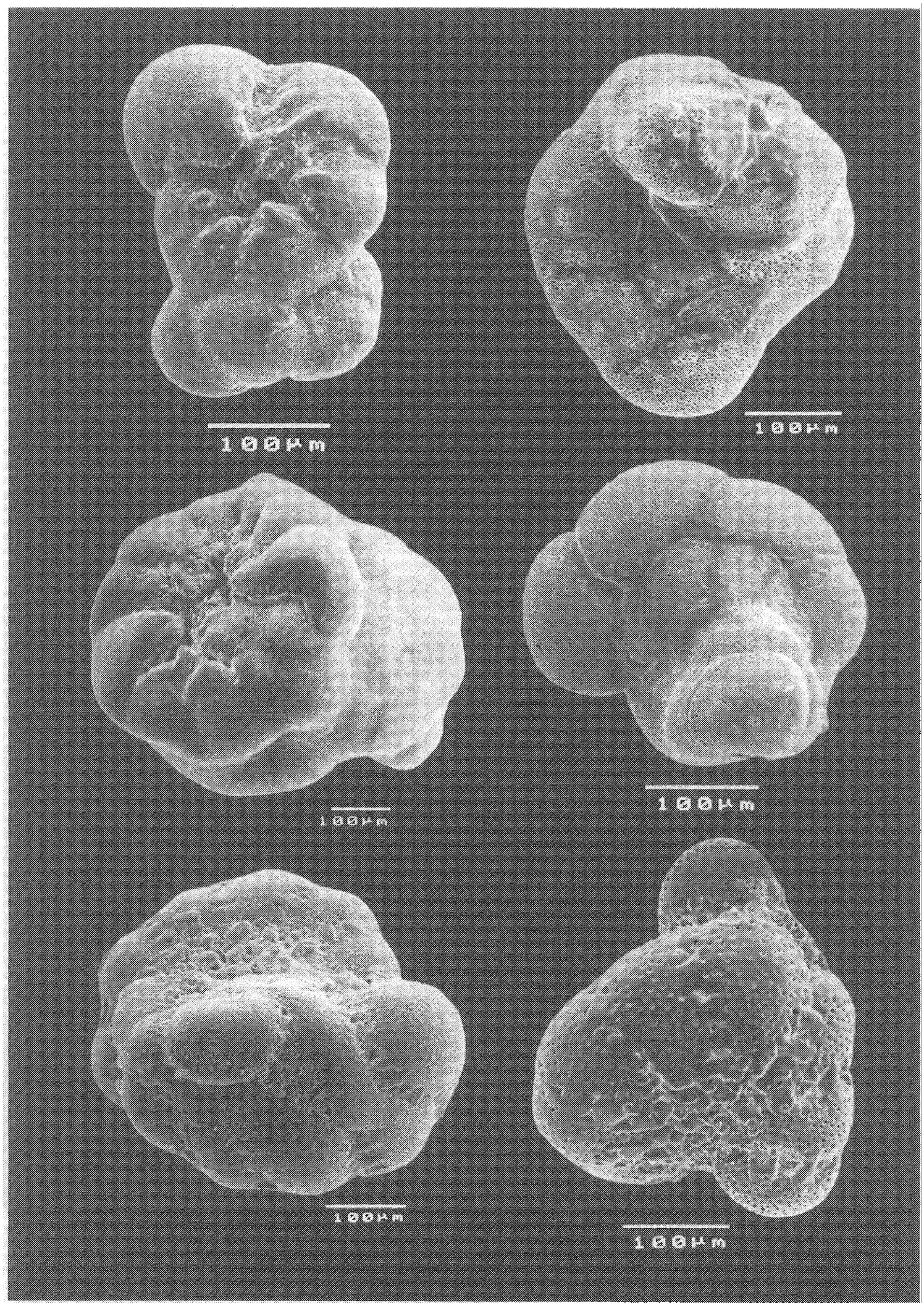

Plate 1. Deformed individuals of the main foraminiferal species. Upper and middle rows: *A. beccarii*, lower row: *E. oceanensis*.

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Appendix 1. Faunal reference list

Porcellaneous species:
Quinqueloculina jugosa Cushman, 1944
Quinqueloculina lata Terquem, 1876
Quinqueloculina seminula (Linné) = *Serpula seminulum* Linné, 1758
Quinqueloculina sp.1
Quinqueloculina sp.2

Hyaline species:
Ammonia beccarii (Linné) = *Nautilis beccarii* Linné, 1758
Brizalina difformis (Williamson) = *Textularia variabilis* var. *difformis* Williamson, 1858
Brizalina spathulata (Williamson) = *Textularia variabilis* var. *spathulata* Williamson, 1858
Brizalina variabilis (Williamson) = *Textularia variabilis* Williamson, 1858
Cassidulina carinata Silvestri, 1896
Cassidulina obtusa Williamson, 1858
Cibicides lobatulus (Walker & Jacob) = *Nautilus lobatulus* Walker & Jacob, 1798
Elphidium crispum (Linné) = *Nautilus crispus* Linné, 1758
Elphidium oceanensis (d'Orbigny) = *Polystomella oceanensis* d'Orbigny, 1826
Elphidium williamsoni Haynes, 1973
Gavelinopsis praegeri (Heron-Allen & Earland) = *Discorbina praegeri* Heron Allen & Earland, 1913
Haynesina depressula (Walker & Jacob) = *Nautilus depressulus* Walker & Jacob, 1798
Haynesina germanica (Ehrenberg) = *Nonion germanicum* Ehrenberg, 1840
Lenticulina inortatus (d'Orbigny) = *Robulina inortata* d'Orbigny, 1846
Rosalina irregularis (Rhumbler) = *Discorbina irregularis* Rhumbler, 1906

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Nutrient inputs to the Irish Sea: temporal and spatial perspectives

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Key words: Irish Sea, dissolved available inorganic nitrogen, dissolved available inorganic phosphorus, nutrient–salinity ratios, sediment remobilisation

Abstract

Data from five key regions are presented to produce thematic plots of winter nutrient (dissolved available inorganic nitrogen and phosphorus) concentrations for the Irish Sea. It is suggested that riverine discharges are the biggest contributor of nutrients to the system, however, anthropogenic point source discharges are also major contributors of these nutrient salts in the eastern Irish Sea. Comparison of the thematic maps produced in this study are made with similar maps produced almost a decade previously, the comparisons show a great many similarities between the two studies. However, waters in the north-eastern Irish Sea show that dissolved available inorganic phosphorus (DAIP) appears to have decreased in concentration in recent years whilst no such change is seen with respect to dissolved available inorganic nitrogen (DAIN). The use of nutrient-salinity regressions indicate that during winter 1997 anthropogenic discharges along the Cumbria coast combined with remobilisation of sedimented material was a major contributing factor of nutrient salts to the system.

Introduction

The Irish Sea is a semi-enclosed waterbody connected to the Atlantic Ocean via the North and St George's Channels (Fig. 1). In general terms, the Sea can be split into two. Waters to the west of the Isle of Man are generally more saline, deeper and have shorter residence times whilst those to the east are shallower, are much more influenced by freshwater inputs and have higher residence times. Residual surface water currents are northerly and residence times have been estimated (see Dickson & Boelens, 1988) between 1.4 months (North Channel) and 7 months (Liverpool Bay). Research into the nutrient salt concentrations of the Irish Sea extend back to the 1950s (Slinn & Eastham, 1984; Allen et al., 1998). These long-term data series suggest an approximate doubling of soluble inorganic nitrogen and phosphorus over this time interval (Allen et al., 1998).

In more recent years, regular monitoring of the Irish Sea via government institutions and university departments has allowed nutrient salt concentrations to be mapped for parts of the Irish Sea. However, only one study has as yet tried to map nutrient salt concentrations across the Irish Sea as a whole (NORSAP, 1992). Data gathered during the winter of 1991 by the NORSAP partners allowed thematic maps to be produced for nitrate and phosphate. This data set was, however, based upon only one year's sampling results and as such does not take into account any interannual variation. Results presented for the present study represent data gathered for five distinct regions in the Irish Sea, Belfast Lough (Department of Agriculture-Northern Ireland), West of the Isle of Man (University of Liverpool), Cumbrian Coast and Liverpool Bay (University of Liverpool/Environment Agency U.K.) and the southern Irish coast (FRC-Marine Institute). Each data set comprises data collected over a 3–6 year time period during the months of January–March (inclusive). Such an exercise allows for a greater understanding of interannual variations in nutrient salt concentrations across the Irish Sea during the winter nutrient maximum.

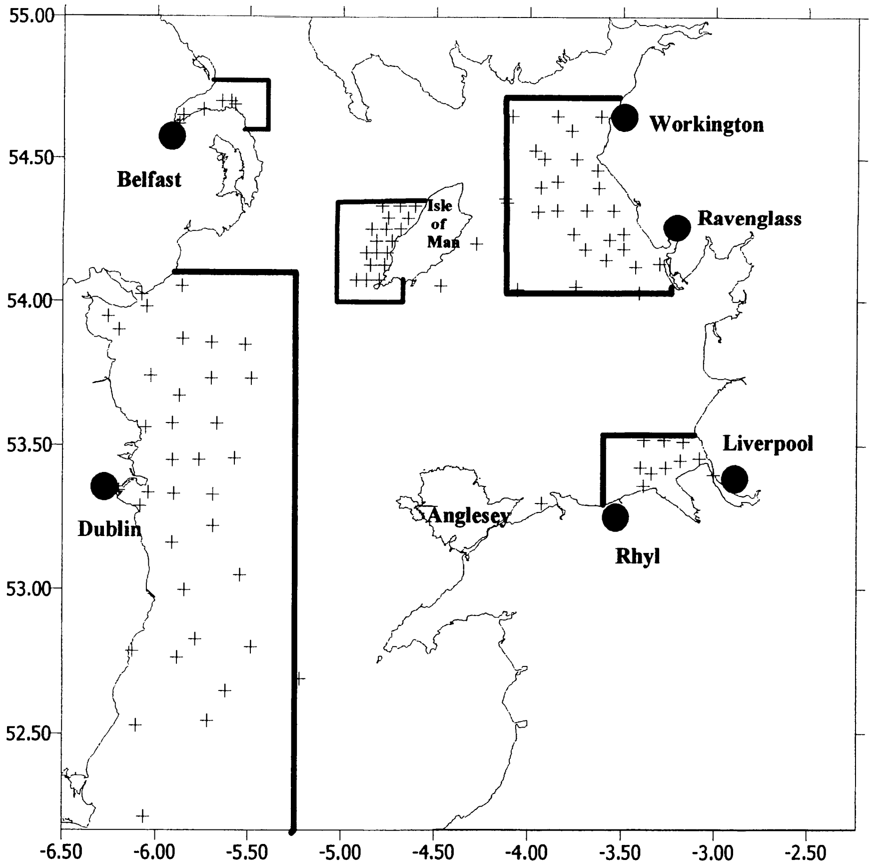

Figure 1. Map showing the five study areas and approximate positions of sample locations.

This paper aims to use several long term data sets to develop thematic maps of dissolved available inorganic nitrogen (NO_2+NO_3+NH_4) and phosphorus (DAIN and DAIP, respectively) for several regions in the Irish Sea. Each of these regions has quite different nutrient salt regimes and there are several different mechanisms by which they receive these nutrients. Data from the southern Irish coastline are also provided to give an indication of nutrient concentrations entering the Irish Sea via the St George's channel. By paying particular attention to nutrient–salinity relationships it was possible to gain some understanding of the origins of nutrient salts entering the Irish Sea.

Methods

Data collected for the Cumbria coast surveys were collected during February 1996, March 1997, January 1998 and January 1999 (for methods see Kennington et al., 1999a, 1999b). Liverpool Bay data were collected during January 1997, February 1998 and February 1999 and methodologies are outlined in Kennington et al. (1999b). Data for the west coast of the Isle of Man result from a long-term monitoring programme based at Port Erin Marine Laboratory (PEML), pooled data being used to give an average for each of the sampling sites between the months of January and March for 1998–2000 (inclusive) and methodologies are the same as described in Kennington et al. (1999b). Data for the East Coast of the Republic of Ireland are from approximately five years earlier than those from the other four regions and represents pooled data collected between January and March for the years 1991–1996 (inclusive). Belfast Lough data also represent pooled data collected between the months of January and March for the years 1995–1997 (inclusive) and 1999. Data for the East Coast of the Republic of Ireland and Belfast Lough used in the present study are those of the Fisheries Research Council (Marine Institute of Southern Ireland) and of the Department of the Environment (Northern Ireland) respectively and methodologies conform to standard sampling protocols used by those organisations.

Statistical analysis was performed using Microsoft Excel and Grapher version 2 (Golden Software inc.). The thematic plots were created using Surfer version 6 (Golden Software inc.). The contour diagrams produced are determined using the Kriging method and grids are blanked using a detailed land boundary. This process extrapolates data for areas where no samples were taken (see Fig. 1).

Results

Thematic maps of DAIN & DAIP

Thematic maps of winter DAIN & DAIP concentrations are plotted in Figures 2 and 3. These maps represent average winter concentrations for the five key regions in the study. It can be seen that there is a clear east-west divide in coastal concentrations of these nutrient salts in the Irish Sea. Each of the five study regions is explained separately below. Data from the Southern Irish Coast is provided for comparison only since these data represent a different time span than for the other regions.

Southern Irish Coast

Waters entering the Irish Sea via the St George's Channel (between 1991 and 1996) had average winter DAIN concentrations of less than 10 $\mu \mathrm{ml}^{-1}$. Coastal concentrations were generally less than 15 $\mu \mathrm{ml}^{-1}$ with the exception of Dundalk Bay where average concentrations reached 25–30 $\mu \mathrm{ml}^{-1}$. A similar distribution was seen with regard to DAIP. Average winter DAIP concentrations of waters entering the Irish Sea were less than 0.6 $\mu \mathrm{ml}^{-1}$. Concentrations increased towards the coast reflecting the added inputs from riverine sources but did not exceed 1 $\mu \mathrm{ml}^{-1}$.

Belfast Lough

Waters entering Belfast Lough during winter are greatly affected by the River Lagan. Average winter DAIN concentrations at the low salinity end members exceed 100 $\mu \mathrm{ml}^{-1}$, these high concentrations becoming diluted fairly readily via mixing with higher salinity waters so that at the mouth of the Lough average winter DAIN concentrations are less than 20 $\mu \mathrm{ml}^{-1}$. Concentrations of DAIP are also greatly affected by the River Lagan with average winter concentrations exceeding 3 $\mu \mathrm{ml}^{-1}$ in the low salinity waters. Average concentrations at the mouth of the Lough are less than 0.5 $\mu \mathrm{ml}^{-1}$, similar to concentrations entering the Irish Sea from the south.

West Coast of the Isle of Man

Waters in the mid Irish Sea show the lowest concentrations of all five data sets. Average winter DAIN concentrations were slightly affected by run-off from the Isle of Man where coastal concentrations reached 8 $\mu \mathrm{ml}^{-1}$. DAIP concentrations were generally less than 0.7 $\mu \mathrm{ml}^{-1}$ across the study area. The average winter concentrations of both DAIN and DAIP at this location show that the offshore Irish Sea waters are very similar in concentration to those entering the Irish Sea via the St George's channel.

Cumbria Coast

Average winter Concentrations of DAIN increase toward the Solway Firth. These concentrations exceed 30 $\mu \mathrm{ml}^{-1}$ and reflect the high freshwater discharge of the Solway itself. Concentrations remain relatively high along the Cumbria coast where they do not fall below 20 $\mu \mathrm{ml}^{-1}$. Offshore waters have average concentrations of less than 10 $\mu \mathrm{ml}^{-1}$, comparable to levels recorded for waters entering the Irish Sea via the St George's Channel.

Average winter DAIP concentrations also increase in concentration towards the Cumbria coast with concentrations reaching 2 $\mu \mathrm{ml}^{-1}$. Offshore waters once more show concentrations similar to waters entering the Irish Sea from the south (<0.8 $\mu \mathrm{ml}^{-1}$).

Liverpool Bay

Concentrations of both DAIN and DAIP increase across the salinity gradient of Liverpool Bay with highest concentrations found in waters adjacent to the Rivers Mersey and Dee (>60 $\mu \mathrm{ml}^{-1}$ DAIN and >2 $\mu \mathrm{ml}^{-1}$ DAIP, respectively). Offshore waters remain enriched by these rivers with concentrations of both DAIN and DAIP being recorded at more than twice those recorded for waters entering the Irish Sea (~ 20 $\mu \mathrm{ml}^{-1}$ DAIN; ~ 1 $\mu \mathrm{ml}^{-1}$ DAIP).

Conservative vs non-conservative nutrient–salinity ratio

The use of nutrient–salinity plots can indicate whether these nutrients are from riverine origin or otherwise. An idealised relationship between concentrations of dissolved inorganic nutrients and salinity shows a conservative or linear response. In such a scenario, low

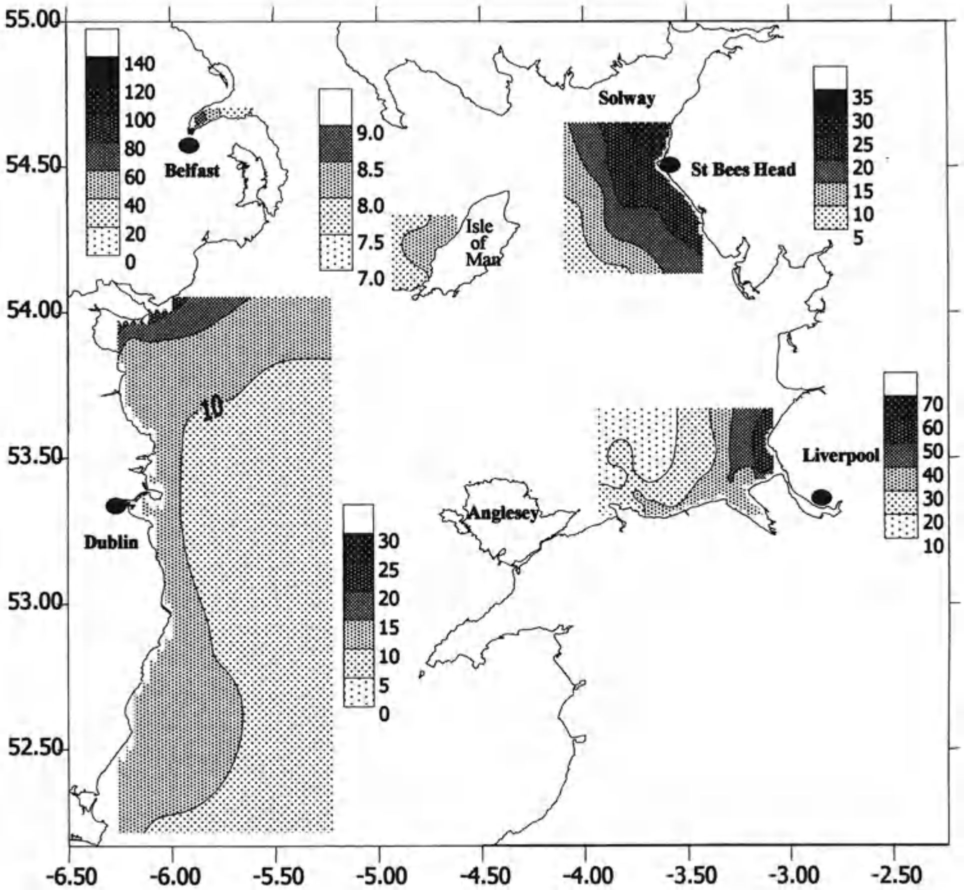

Figure 2. Thematic plot of winter (Jan–March) dissolved available inorganic nitrogen across the five study areas (μml^{-1}).

salinity waters generally have higher nutrient concentrations and higher salinity waters have lower nutrient concentrations. Any deviation from this trend would, therefore, indicate additional inputs of nutrients via processes other than riverine ones (e.g. industrial/municipal discharges, adsorption/desorbtion by suspended matter etc.).

Liverpool Bay and Belfast Lough

Regression analyses between major nutrient salt concentrations and salinity within Liverpool Bay for the years 1997 and 1998 and in Belfast Lough for 1999 are plotted in Figures 4 and 5. It can be seen that a conservative relationship exists between DAIN/DAIP and salinity in both waterbodies. These relationships suggest that nutrients entering Liverpool Bay and Belfast Lough do so mainly via riverine discharges. The relatively high nutrient concentrations entering Belfast Lough reflect industrial point source discharges of both N and P at the mouth of the River Lagan (Service et al., 1996). These discharges occur in low salinity waters and as such do not alter the conservative nutrient–salinity relationships.

Cumbria Coast

Figures 6 and 7 show winter DAIN and DAIP maxima for the years 1996–1999 plotted against salinity. It can be seen that for three of these years (1996, 1998 & 1999) a conservative (linear) relationship exists between DAIN/DAIP and salinity. These relationships suggest that the majority of N and P entering the system are of riverine origin. Plots 6(c) and 7(c) show the DAIN-salinity and DAIP-Salinity relationships for March 1997. It can be seen that the conservative relationships described above are not applicable for this year. These non-conservative responses suggest that riverine loading is not the only major source of DAIN and DAIP to the system. The factors that might cause such a change in the functional responses of DAIN-salinity and DAIP-Salinity include uptake of N and P

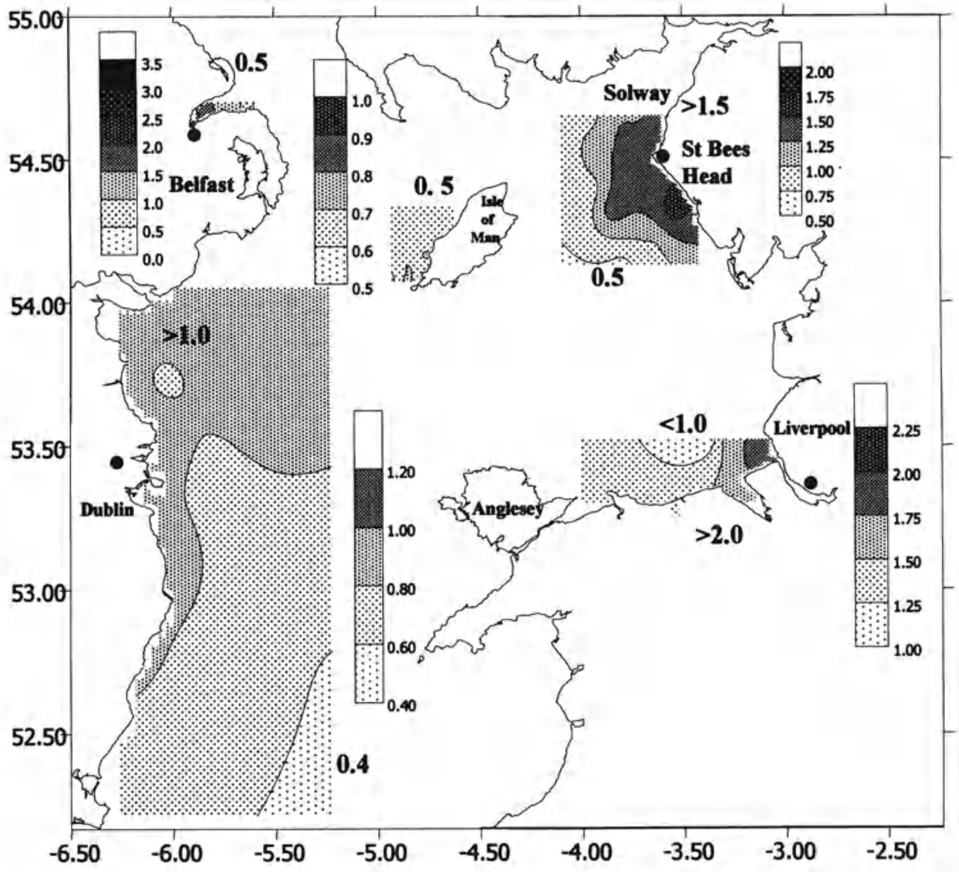

Figure 3. Thematic map of winter (Jan–March) dissolved available inorganic phosphorus across the five study areas (μml^{-1}).

by phytoplankton, atmospheric deposition of nitrogen, remobilisation of marine sediments and point source discharges to the coastal environment.

Discussion

Nutrients entering the Irish Sea do so via several mechanisms. The contribution of riverine run-off is particularly evident for the eastern Irish Sea where the Solway Firth, Mersey and Dee estuaries are the major contributors. Waters in the western Irish Sea also receive riverine sourced nutrients but to a much lesser degree. Furthermore, these nutrients may not reach the higher concentrations found in the east owing to the faster residual water currents and deeper receiving waters, which create a greater dilution for such inputs. These factors, combined with the shorter residence times of waters in the western Irish Sea, aid the dispersal of riverine sourced nutrient inputs. The contribution of both DAIN and DAIP via Belfast Lough is negligible as waters discharging here would soon be incorporated into the northern flowing current exiting the North Channel. Central Irish Sea waters appear to be relatively unaffected by riverine sourced nutrient inputs with concentrations of both DAIN and DAIP being close to those of waters entering the Irish Sea from the south (~10 μml^{-1} DAIN and ~0.4 μml^{-1} DAIP, see Figures 2 and 3). It was mentioned previously that long term nutrient monitoring of mid-Irish Sea waters have reported an approximate doubling of both N and P during the past 40 years (Allen et al., 1998). The results presented for the present study have shown that over a three year average there does not seem to be any major difference between concentration of N and P in mid-Irish Sea waters and those entering the Sea via the St George's Channel. This being so, it calls into question the origin of N and P entering the system. Waters entering via the St George's Channel may not be representative of open Atlantic conditions and increases in concentrations of N and P recorded in the mid-Irish Sea may in fact be as a result

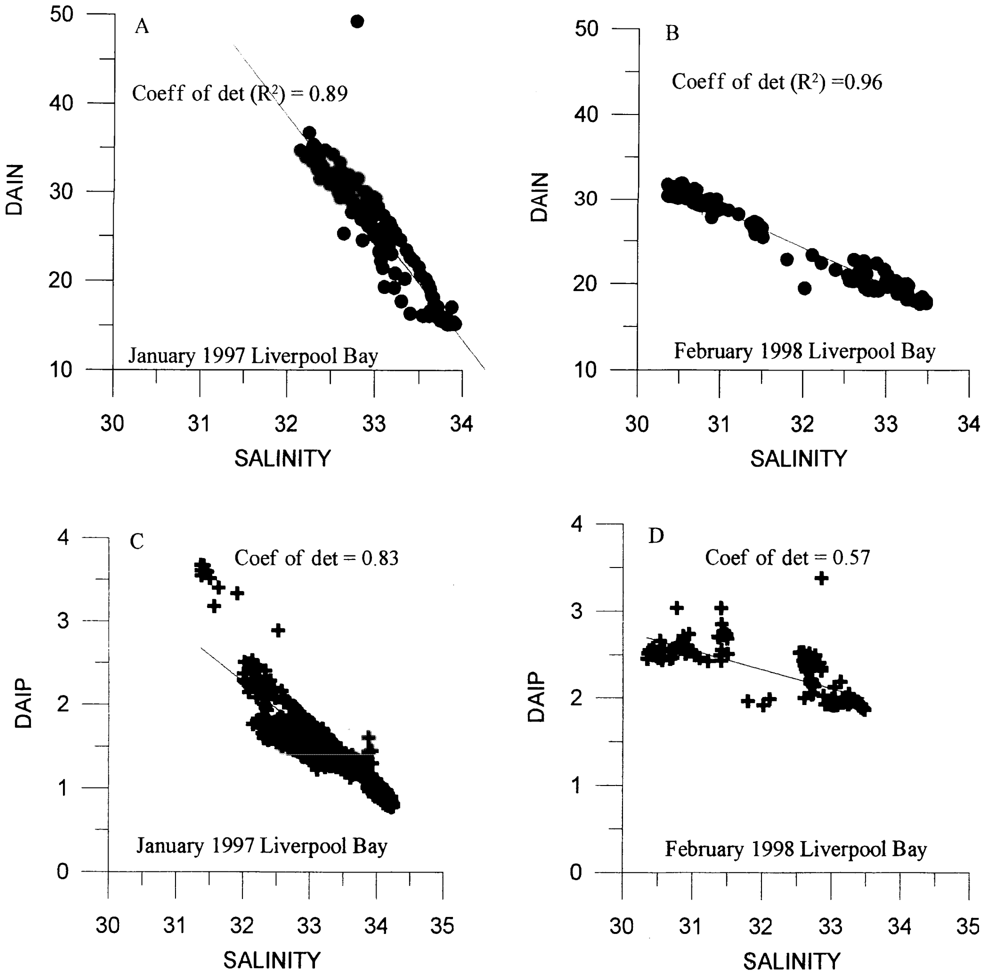

Figure 4. A & B: Liverpool Bay winter surveys 1997–1998 DAIN (μml^{-1}) vs salinity. C & D: Liverpool Bay winter surveys 1997–1998 DAIP (μml^{-1}) vs salinity.

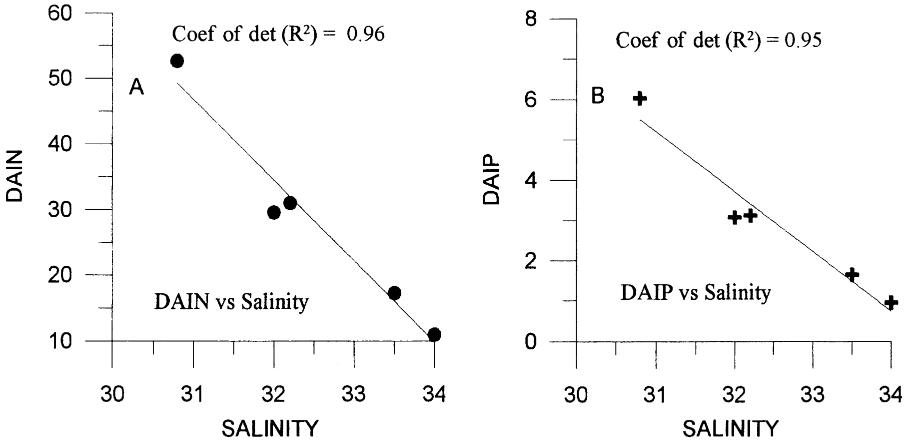

Figure 5. A & B: Belfast Lough Winter Surveys 1999 (μml^{-1}) vs salinity.

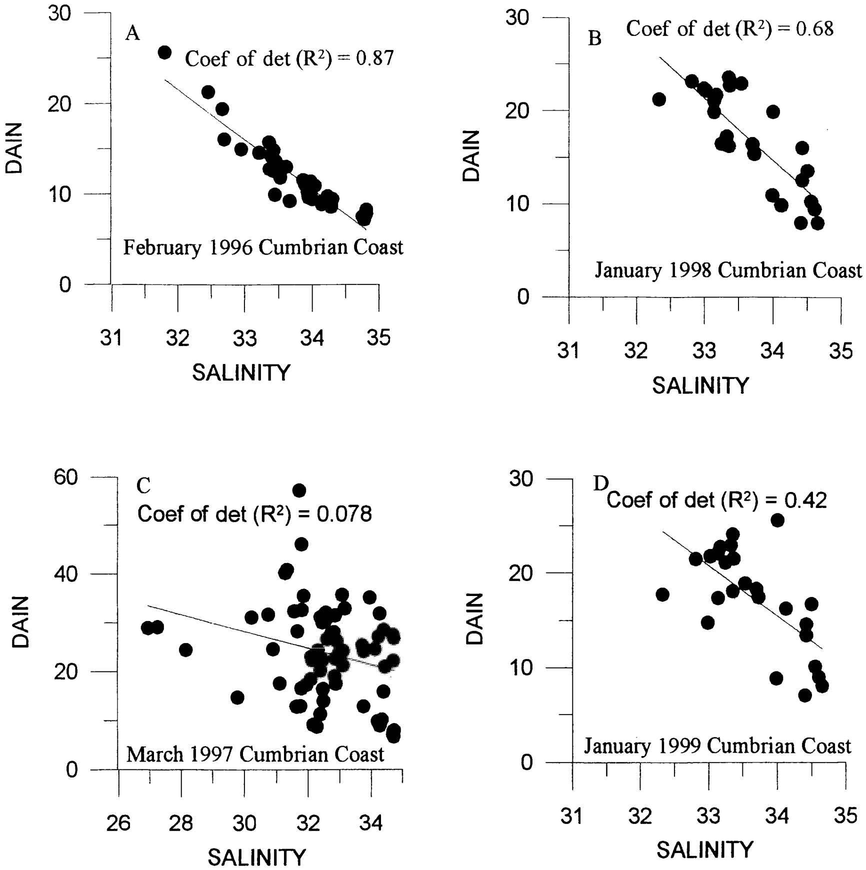

Figure 6. Cumbria Coast Winter Surveys 1996–1999 (inclusive) DAIN (μml^{-1}) vs salinity.

of increased discharges into the Celtic Sea (e.g. from the River Severn) which is subsequently entrained into the northerly flowing waters entering the Irish Sea. However, since only three years data were used in the present study for the analysis of mid-Irish Sea waters it is difficult to make any real comparison with the longer term trends. Concentrations between years have been shown to be highly variable and it is only by observing these results over time-scale of decades that such trends can really be appreciated.

The uptake of N and P by phytoplankton does not seem to have been a major contributing factor for the non-conservative response outlined for the Cumbria coast during 1997. Chlorophyll levels recorded during the survey were still very low (generally less than 1 μg/litre). If uptake of N and P by phytoplankton was the causative mechanism for change in the conservative relationships then there would be a significant decrease in N and P across the study area. Concentrations of DAIN and DAIP during March 1997 were in fact considerably higher than for the other three winter sampling dates (see Kennington et al., 1998).

During winter storms, the potential for remobilisation of marine sediments is intensified. Such storms cause deep water mixing bringing sedimented material back into suspension. This suspended material is often rich in inorganic nitrogen and phosphorus and as such would cause an increase in DAIN and DAIP of the overlying waters. In addition to increases in DAIN and DAIP, such storms could also increase the concentration of dissolved silicate. Silicate enters the marine system either through weathering processes on land and subsequent transport by water or ice, or by dissolution of biogenic silica. Remobilisation of sediments rich in biogenic silica (e.g. as diatom frustules) would allow any silicate present to be prone to dissolution. Figure 8 shows the silicate-salinity relationships along the Cumbria coast for the years 1996–1999. It can be seen that generally there is a good conservative response between these variables for most years. Plot 8 (c) represents the silicate-salinity relationship for 1997

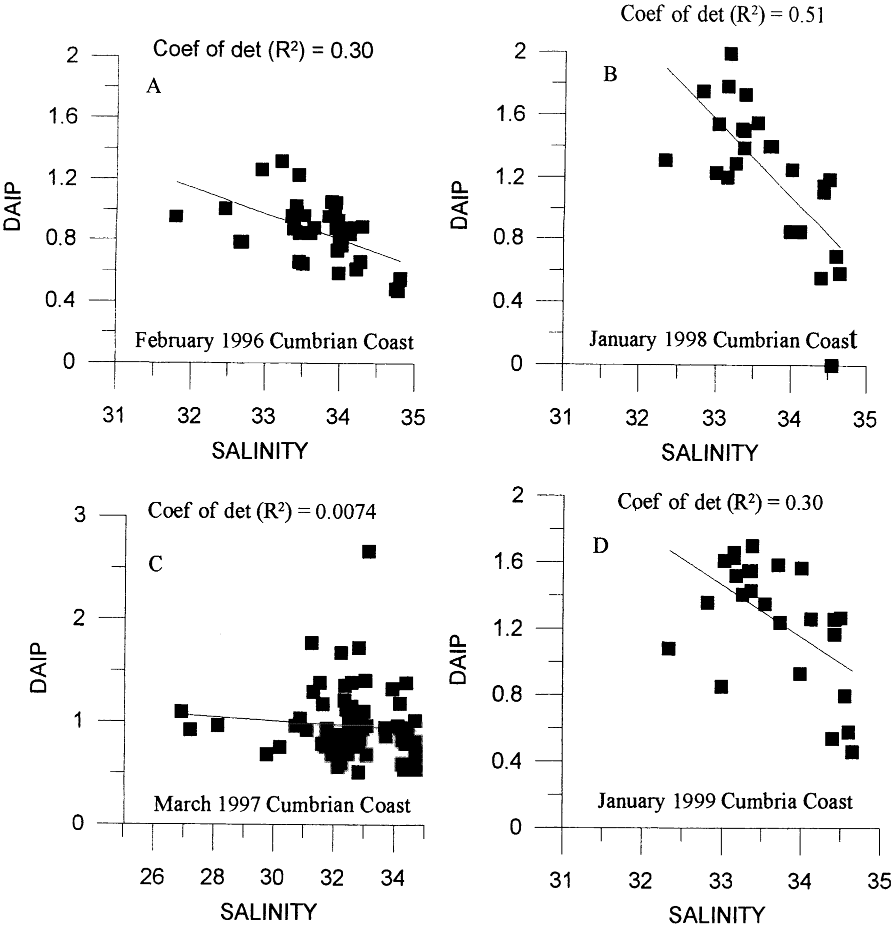

Figure 7. A–D: Cumbria Coast Winter Survey 1996–1999 (inclusive) DAIP (μml^{-1}) vs salinity.

and shows the weakest response of the four years and suggests that there may possibly be added inputs of silicate from non-riverine sources (i.e. remobilisation).

Figure 9 (a–c) shows meteorological data for the Irish Sea for the two months preceding the sampling period. It can be seen from Figure 9 (a) that wind speed increased during February with an average speed of approximately 20 knots and gusts of up to 60 knots prevailing (Fig. 9 (b)). The wind direction at this time was primarily south-westerly (Fig. 9 (c)). Such strong onshore winds may, therefore, have caused a degree of deep water mixing in the weeks prior to the 1997 sampling dates. Wind strengths for the three other years studied have indeed been recorded at strengths equal to and stronger than those for 1997, however, the persistent south-westerly direction for over a month prior to the sampling trip in 1997 combined with these strong winds was unique to 1997 and not found during the other three years studied.

Point source discharges of DAIN and DAIP to the coastal waters could also alter the relationship between these variables and salinity. Such point source discharges along the Cumbria coast include sewage outfall pipes and industrial discharges from Whitehaven and Sellafield. The impact of such discharges directly into higher salinity waters would cause a shift in the DAIN/DAIP-salinity relationship. High concentrations of DAIN were recorded from the Solway Firth and in waters adjacent to Sellafield during 1997 (see Kennington et al., 1998). Similarly highest DAIP concentrations during this time were recorded between Workington and Ravenglass. It seems most likely, therefore, that the contribution of point source discharges along the Cumbria coast combined with possible remobilisation of sediments rich in N, P, and

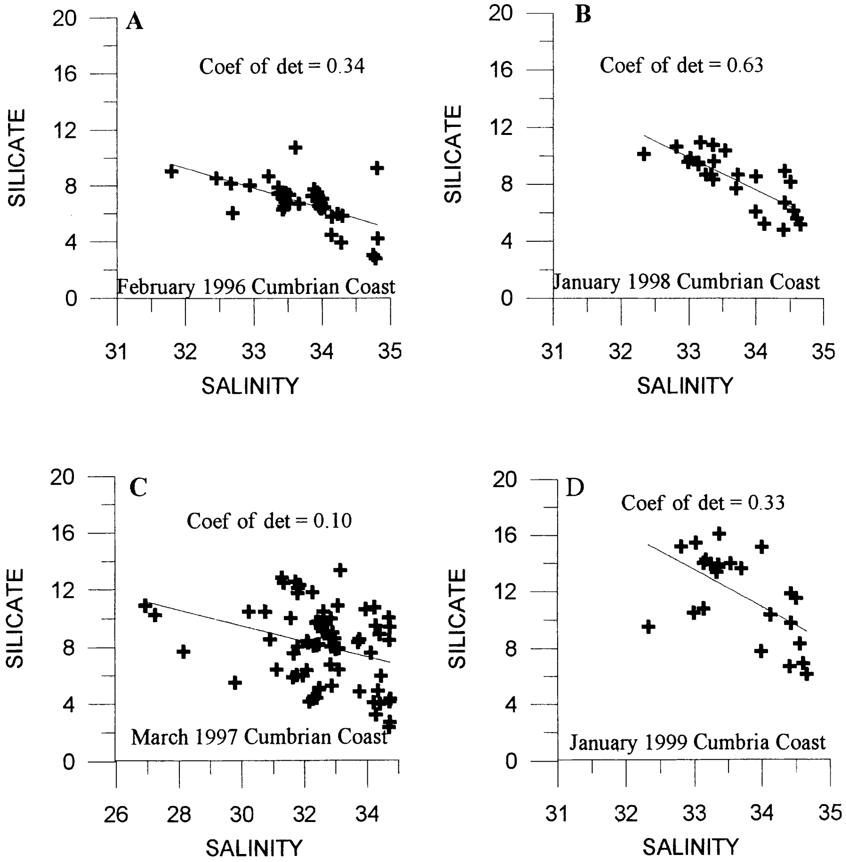

Figure 8. A–D: Cumbria Coast Winter Survey 1996–1999 (inclusive) Silicate (μml^{-1}) vs salinity.

Si are responsible for the non-conservative relationships between major nutrient salts and salinity during 1997.

The contribution of N from atmospheric deposition to the Irish Sea is as yet unquantified. Such studies have been completed for other North Atlantic waters. The contribution of N-nitrogen to the North Sea has been estimated at up 50% of the total riverine N contribution (Simpson, 1994). If such a scenario were true for the Irish Sea then this would still not necessarily remove the conservative relationship between DAIN and salinity. The atmospheric contribution would be relatively even across all coastal waters and atmospheric deposition over land would contribute to the low salinity–high nutrient end members.

Conclusions

Nutrient inputs to the Irish Sea have been shown to be highest in the eastern Irish Sea. Here the influence of riverine and point source discharges into the system is relatively greater than those from the Irish coasts. This, combined with the lower overall volume of the eastern Irish Sea and its longer residence times, indicates that these waters are more prone to enrichment than those to the west. The thematic maps developed in this study are in very close agreement with those produced by the NORSAP (1992) study. One major alteration has been an apparent decrease in DAIP concentrations along the Cumbria coast from a maximum of 4 μml^{-1} in 1991 to a maximum of 2 μml^{-1} between 1996 and 1999. This reduction may reflect the fact that the NORSAP data was collected during only one season (which may have been higher than average) or it may reflect the reduction in anthropogenic inputs from both industry and sewage that have occurred during the intervening years. The concentrations of DAIN on the other hand seems to have shown no such reduction despite a reduction in N to the system via point source discharges to the Irish Sea (U.K. Environment Agency, unpublished data).

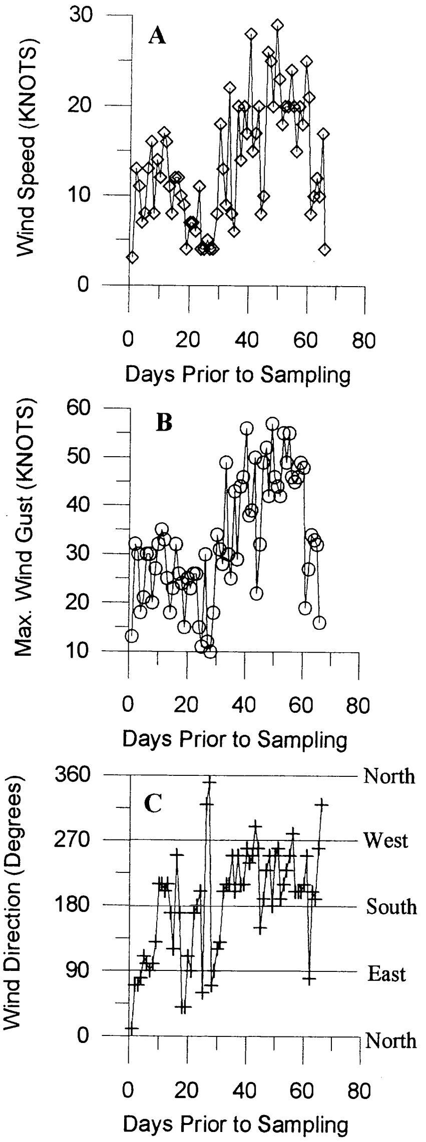

Figure 9. A–C: Meteorological Office (Ronaldsway) data 1997.

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The authors would like to thank Dr Eugene Nixon (Marine Institute-Southern Ireland) and Roslyn Todd (Environment & Heritage Service, Northern Ireland) for the use of the western Irish Sea and Belfast Lough datasets. Ronaldsway Meteorological Office are also thanked for making available the meteorological data used in this study. The crew and research staff of the R.V. Roagan and R.V. Coastal Guardian are thanked for the winter trips up and down the eastern Irish Sea coast in often severe weather conditions.

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Nutrient-enhanced productivity in the northern Gulf of Mexico: past, present and future

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Key words: eutrophication, hypoxia, Gulf of Mexico, Mississippi River, nutrient-enhanced productivity, river–ocean interactions

Abstract

Nutrient over-enrichment in many areas around the world is having pervasive ecological effects on coastal ecosystems. These effects include reduced dissolved oxygen in aquatic systems and subsequent impacts on living resources. The largest zone of oxygen-depleted coastal waters in the United States, and the entire western Atlantic Ocean, is found in the northern Gulf of Mexico on the Louisiana/Texas continental shelf influenced by the freshwater discharge and nutrient load of the Mississippi River system. The mid-summer bottom areal extent of hypoxic waters (<2 mg l^{-1} O_2) in 1985–1992 averaged 8000 to 9000 km^2 but increased to up to 16 000 to 20 700 km^2 in 1993–2001. The Mississippi River system is the dominant source of fresh water and nutrients to the northern Gulf of Mexico. Mississippi River nutrient concentrations and loading to the adjacent continental shelf have changed in the last half of the 20th century. The average annual nitrate concentration doubled, and the mean silicate concentration was reduced by 50%. There is no doubt that the average concentration and flux of nitrogen (per unit volume discharge) increased from the 1950s to 1980s, especially in the spring. There is considerable evidence that nutrient-enhanced primary production in the northern Gulf of Mexico is causally related to the oxygen depletion in the lower water column. Evidence from long-term data sets and the sedimentary record demonstrate that historic increases in riverine dissolved inorganic nitrogen concentration and loads over the last 50 years are highly correlated with indicators of increased productivity in the overlying water column, i.e. eutrophication of the continental shelf waters, and subsequent worsening of oxygen stress in the bottom waters. Evidence associates increased coastal ocean productivity and worsening oxygen depletion with changes in landscape use and nutrient management that resulted in nutrient enrichment of receiving waters. A steady-state model, calibrated to different observed summer conditions, was used to assess the response of the system to reductions in nutrient inputs. A reduction in surface layer chlorophyll and an increase in lower layer dissolved oxygen resulted from a reduction of either nitrogen or phosphorus loading, with the response being greater for nitrogen reductions.

Introduction

A variety of environmental changes may result in the increased accumulation of organic matter in a marine system (= eutrophication, as defined by Nixon, 1995) with the most common single factor being an increase in the amount of nitrogen and phosphorus being delivered to marine waters. With an increase in the world population, a focusing of that populace in coastal regions, agricultural expansion in major river basins, and increased energy consumption, eutrophication is becoming a major environmental problem in coastal waters throughout the world. There are clear signals that humans have altered the global cycles of nitrogen and phosphorus over large regions and increased the mobility and availability of these nutrients to marine ecosystems (Peierls et al., 1991; Howarth et al., 1995, 1996; Vitousek et al., 1997; Howarth, 1998; Caraco & Cole, 1999; Bennett et al., 2001). Human-controlled inputs derive from the increase in human populations and their activities, particularly from the application of nitrogen and phosphorus fertilizers, nitrogen fixation by leguminous crops, and atmospheric deposition of oxidized nitrogen from fossil-fuel combustion. Changes in the relative proportions of these nutrients may exacerbate eutrophication, favor noxious algal blooms and aggravate conditions of oxygen depletion (Officer & Ryther, 1980; Conley et al., 1993; Justic et al., 1995a,b; Turner et al., 1998). Global patterns of temporal change toward increased loads of nutrients from watersheds and worsening coastal water quality (e.g. Justic et al., 1987; Andersson & Rydberg, 1988; Cooper & Brush, 1991; Hickel et al., 1993; Diaz & Rosenberg, 1995) is exemplified by the Mississippi River watershed/Gulf of Mexico coastal ecosystem.

The Mississippi River system dominates the flux of fresh water and associated dissolved and particulate materials on the central northern Gulf of Mexico shelf. The waters of the continental shelf adjacent to the Mississippi River are affected on varying spatial and temporal scales by a discharge that integrates runoff from 41% of the lower 48 United States. The Mississippi and Atchafalaya Rivers combined (the two discharges for the system) contribute 580 $km^3\ y^{-1}$ of fresh water to the Gulf of Mexico along with sediment yields of 210×10^6 t y^{-1}, 1.6×10^6 t y^{-1} nitrogen, of which 0.95×10^6 t is nitrate and 0.58×10^6 t is organic nitrogen, 0.1×10^6 t y^{-1} phosphorus and 2.1×10^6 t y^{-1} silica (Milliman & Meade, 1983; Meade, 1995; Goolsby et al., 1999). The ecosystem adjacent to the discharges of the Mississippi River system, unlike many estuaries and confined seas, is an open continental shelf system. There is daily, weekly and seasonal variability in currents and stratification on the shelf and, therefore, no simple description of the couplings between nutrient delivery, carbon production in surface waters and delivery to and cycling in bottom waters. There are, however, multiple lines of evidence to implicate changes in riverine nutrient loads with overall primary and secondary production, carbon accumulation at the seabed, and low oxygen conditions on the shelf. The seasonally severe hypoxic zone that develops at the terminus of the Mississippi River system is among the largest in the world's coastal ocean, reaching up to 20 700 km^2 of bottom water in mid-summer (Rabalais & Turner, 2001).

A series of experiments, empirical relationships, monthly oxygen and carbon budgets, time series analyses, models and comparisons with other regions of the world's coastal ocean indicate a close coupling between Mississippi river-borne nutrients, primary and net productivity, vertical carbon flux and hypoxia on time scales as short as days and as long as decades and centuries. In this paper, we synthesize these various aspects of nutrient-enhanced productivity and hypoxia on the northern Gulf of Mexico continental shelf. We identify the linkages of nutrient load to productivity and oxygen stress, and how these relationships have changed over the last century and a half, most dramatically since the 1950s. We present data mostly from published results that provide the appropriate details of collection and analysis methods. We also present scenarios of nutrient load reductions and how those reductions might influence coastal water quality.

Hypoxia, characteristics and causes

The mid-summer bottom areal extent of hypoxic waters (<2 mg l^{-1} O_2, or ppm) in 1985–1992 averaged 8000 to 9000 km^2 but increased to 16 000 to 20 700 km^2 in 1993–2001 (Rabalais & Turner, 2001). The estimated extent was 12 500 km^2 in mid-summer of 1998, and 4400 km^2 in 2000. There is variability in the mid-summer extent of hypoxia due to physical conditions at the time of sampling, for example the reduced mid-summer size in 1997 and 1998 due to the passage of a hurricane on the southeastern portion of the study area and the current regime, respectively. A compilation of fifteen mid-summer surveys (1985–1999) demonstrates that the frequency of occurrence of hypoxia is higher to the west of the discharges of the Mississippi and Atchafalaya Rivers in a down-current direction from their influence (Fig. 1).

Critically depressed dissolved oxygen concentrations occur below the pycnocline along transect C from as early as late February through early October and nearly continuously from mid-May through mid-September. In March, April and May, hypoxia tends to be patchy and ephemeral; it is most widespread, persistent, and severe in June, July and August (Rabalais et al., 1999). The low oxygen water mass on the bottom during peak development in the summer changes configuration in response to winds, currents

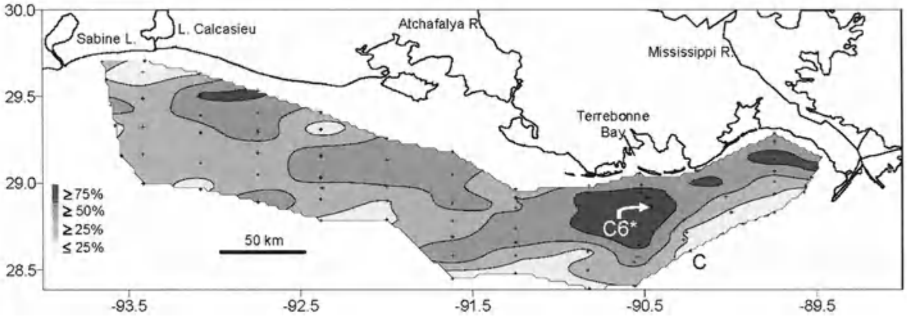

Figure 1. Distribution of frequency of occurrence of mid-summer bottom-water hypoxia over the 60–80-station grid from 1985 to 1999 (derived from Rabalais, Turner & Wiseman, various published and unpublished data). Transect C and station C6*, which incorporates data from C6, C6A and C6B, are identified (from Rabalais & Turner, 2001).

and tidal advection. The persistence of extensive and severe hypoxia into September and October depends primarily on the breakdown of stratification by winds from either tropical storm activity or passage of cold fronts. Hypoxia is rare in the late fall and winter. Hypoxia occurs not only at the bottom near the sediments, but well up into the water column. Depending on the depth of the water and the location of the pycnocline(s), hypoxia may encompass from 10% to over 80% of the total water column, but normally only 20–50%. Anoxic bottom waters can occur, along with the release of toxic hydrogen sulfide from the sediments.

Two principal factors lead to the high productivity and the subsequent development and maintenance of hypoxia on the Louisiana continental shelf – a physically stratified water column and decomposition of photosynthetically-produced organic matter fluxed to the bottom. The physics of the system defines where the biological processes of carbon production, flux and respiration that lead to oxygen depletion can occur, and the seasonal and spatial distribution of nutrients sets the bounds for those processes. The high freshwater discharge, general circulation patterns of the Louisiana shelf and the presence of the Louisiana coastal current dictate a stratified system for much of the year, interrupted on occasion by wind-mixing events, notably tropical storms and winter cold fronts. The relative influence of the physics and the biological processes varies across the broad region where hypoxia occurs and over an annual cycle. The physical features of the system and the biological processes cannot be separated from each other and must be considered within the overall context of a large river interacting with a coastal sea.

River discharge

Of the rivers and streams between Galveston Bay (Texas) and the Mississippi River delta, i.e. those most likely to influence the zone of hypoxia, the combined flows of the Mississippi and Atchafalaya Rivers account for 96% of the annual freshwater discharge and 98.5% of the total nitrogen load (calculated from U.S. Geological Survey streamflow data for 37 U.S. streams discharging into the Gulf of Mexico; Dunn, 1996). A similar calculation for annual total phosphorus load gives 98% for the relative contribution of the Mississippi and Atchafalaya Rivers. The relative contribution of direct atmospheric deposition was estimated by Goolsby et al. (1999), for an area twice the size of the hypoxic zone, to be 1% of that contributed by the discharge of the Mississippi and Atchafalaya Rivers. Sources of nutrients from groundwater discharge and upwelling are presumed to be minimal based on geologic and physical features of the system, but data on exchange of nutrients from these potential sources are limited (Rabalais et al., 1999).

The combined long-term average annual discharge for the Mississippi and Atchafalaya Rivers to the Gulf of Mexico is 19 920 $m^3\ s^{-1}$ (1930–1997 period) (Bratkovich et al., 1994; Goolsby et al., 1999). The long-term peak flow occurs in March, April and May, and the long-term low flow is in summer and early fall. Although flow is reduced in summer, large-scale circulation patterns facilitate the retention of the fresh water on the shelf (Rabalais et al., 1999). There is significant interannual variability in discharge, but the long-term average discharge for the lower Mississippi River is remarkably stable near 14 000 $m^3\ s^{-1}$ (Turner & Rabalais, 1991; Bratkovich et al., 1994).

The discharge of the Atchafalaya increased during the course of the record (1900–1992) examined by Bratkovich et al. (1994), and the trend was associated with the tendency of the river to increasingly capture flow from the Mississippi River proper (up to the 1977 Congressional mandate for a 30% diversion). Less obvious is a statistically significant and increasing trend in the Mississippi River discharge for 1900–1992 as measured at Tarbert Landing (Bratkovich et al., 1994). It appears to be due to a tendency for increasing discharge in September through December. This period, however, is much less important in the coastal ocean than spring and summer in the timing of important biological processes that lead to the development of hypoxia or the physical processes important in its maintenance. If a longer period of annual discharge were considered, e.g. for the early 1800s to present, the trends since the 1950s are obvious but are concealed within high interannual variability and no long-term change over a century and a half (Rabalais et al., 1999).

Nutrient flux and ratios, current and historical changes

Three nutrients in various forms are important for freshwater and marine phytoplankton growth and production. Nitrogen is considered a more dominant influence than phosphorus in estuarine and marine communities (e.g. Valiela, 1984; D'Elia et al., 1986; Harris, 1986), but not all coastal systems are nitrogen limited (e.g. the Huanghe in China, Turner et al., 1990). Diatoms require silicon to build their cell walls (frustules), and are the dominant biomass component of many marine and estuarine phytoplankton communities, particularly in the spring. If silica is limited, other non-siliceous forms, such as dinoflagellates or cyanobacteria, may become proportionally more important in the phytoplankton community. The concentration and relative proportion of these three nutrients to each other are important in phytoplankton production and composition, and subsequent effects on food webs, energy flow and trophodynamics (Dortch & Whitledge, 1992; Justic et al., 1995a,b; Turner et al., 1998).

Several researchers have documented changes in Mississippi River nutrient concentration and flux over varying periods (Smith et al., 1987; Turner & Rabalais, 1991, 1994a,b; Justic et al., 1995a,b; Meade, 1995; Rabalais et al., 1996; Goolsby et al., 1999; Goolsby, 2000). Differences in results from these studies come from analysis of different time periods, different statistical methods, and different calculations of flux. There are also differences depending on whether the concentration of a constituent in the lower River as it approaches the Gulf or its flux to the Gulf is under consideration. Results generally concur that Mississippi River nutrient concentrations and loading to the adjacent continental shelf have changed dramatically this century, with an acceleration of these changes in the last half of the 20th century depending on the constituent of concern.

Turner & Rabalais (1991) examined water quality data for four lower Mississippi River stations for dissolved inorganic nitrogen (as nitrate), phosphorus (as total phosphorus) and silicon (as silicate). The mean annual concentration of nitrate was approximately the same in 1905–1906 and 1933–1934 as in the 1950s, but doubled from the 1950s to 1990s. The increase in total nitrogen is almost entirely due to changes in nitrate concentration. The mean annual concentration of silicate was approximately the same in 1905–1906 as in the early 1950s, then it declined by 50%. Concentrations of nitrate and silicate appear to have stabilized, but trends are masked by increased variability since the 1980s. There are no substantial records of total phosphorus concentration in the lower Mississippi River before 1973 and subsequent values vary greatly among years. Goolsby et al. (1999) found no long-term trend in orthophosphate or total P from 1973 to 1996. Turner & Rabalais (1991) noted a trend for increase in orthophosphate that was masked within extreme variability in the data. Justic et al. (1995b) applied a linear least-squares regression on the 1973–1987 total P data and estimated ($p<0.01$) that the total P concentration increased two fold between 1960–1962 and 1981–1987. This result was corroborated by comparisons of recent measurements (Rabalais et al., unpubl. data) and historical data from coastal waters adjacent to the Mississippi River discharge (Thomas & Simmons, 1960).

The annual flux of nitrate to the Gulf from the Mississippi River Basin also increased significantly during the period 1955–1999 (Goolsby et al., 1999, 2000). The increase in nitrate flux can be attributed partially to an increase in precipitation and streamflow; the average annual streamflow during 1980–1999 was about 30% higher than during 1955–1970. Another reason for the increase in flux is the doubling in the average concentration of nitrate in the lower Mississippi River (Turner & Rabalais, 1991). On a seasonal basis, the

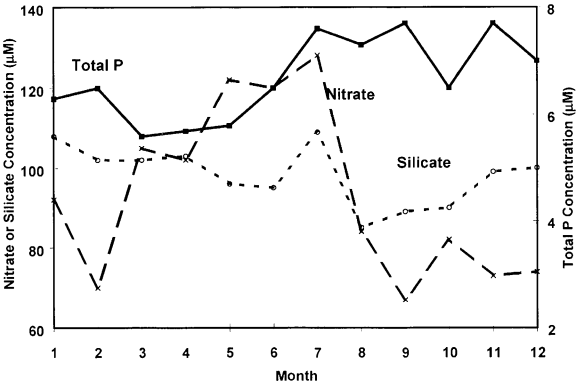

Figure 2. Monthly variation in average (1975–1985) nitrate, silicate and total phosphorus concentrations in the Mississippi River at St. Francisville, Louisiana (redrawn from Turner & Rabalais, 1991).

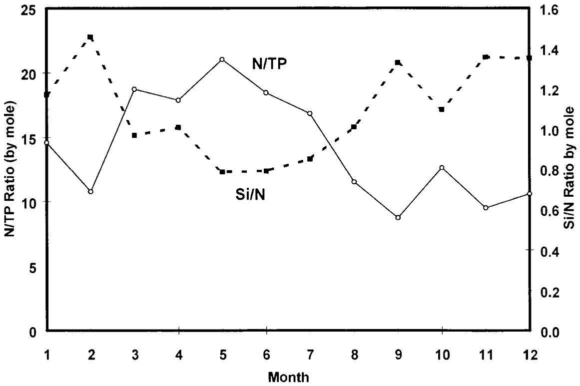

Figure 3. Monthly variation in average (1975–1985) nutrient ratios in the Mississippi River at St. Francisville, Louisiana (drawn from data in Turner & Rabalais, 1991).

period of highest nitrate flux to the Gulf is usually in the spring and early summer, preceding the peak in development of hypoxia in the Gulf.

From 1977 to 1994, the total nitrogen pool averaged 59% nitrate and 37% organic nitrogen. The remaining 4% was ammonium (3%), nitrite (1%) and unidentified dissolved organic nitrogen molecules (Turner & Rabalais, 1991). The importance of dissolved forms other than nitrate and the dissolved organic and particulate organic nitrogen component are not being dismissed in our following discussions, but less is known about their relative contribution of the total flux over a longer-time frame and the related biological processes on the adjacent shelf. Studies from the Mississippi River plume support the view of an active microbial population capable of utilization of organic nitrogen and rapid rates of regeneration of inorganic nutrients (Gardner et al., 1994, 1997; Pakulski et al., 1995; Bode & Dortch, 1996).

The seasonal patterns in nitrate and silicate concentration have also changed during this century. There was no pronounced peak in nitrate concentration earlier this century, whereas there was a spring peak from 1975 to 1985 (Turner & Rabalais, 1991). A seasonal summer–fall maximum in silicate concentration, in contrast, is no longer evident. Currently, there is nearly a two-fold difference in nitrate concentration over the course of the year (Turner & Rabalais, 1991), but only small annual variations in the silicate and total phosphorus concentration (Fig. 2).

The silicate:nitrate ratios have changed as the concentrations varied (Turner & Rabalais, 1991). The Si:N atomic ratio was approximately 4:1 at the beginning of this century, dropped to 3:1 in 1950 and then rose to approximately 4.5:1 during the next 10 years, before plummeting to 1:1 in the 1980s. The ratio appears stable at 1:1 through 1997 with little variation (Turner et al., 1998). The Si:P ratio decreased from 40 to 14, and the N:P ratio increased from 9 to 15. By applying the Redfield ratio as a criterion for stoichiometric nutrient balance, one can distinguish between P-deficient, N-deficient, and Si-deficient waters, and those having a well balanced nutrient composition. The average atomic ratios of N:Si, N:P and Si:P are currently 1.1, 15 and 14, respectively, and closely approximate those of Redfield (1958) of 16:16:1, N:Si:P (Justic et al., 1995a, b). The nutrient ratios for the Mississippi River on an annual basis (1981–1987 database) show an almost perfect coincidence with the Redfield ratio and suggest a balanced nutrient composition. Although the concentrations of nitrate and silicate currently average near 100 μM on an annual basis, there is much variability about this average. Total P averages 7.4 μM at the mouth of the Mississippi River and is lowest in the spring. Thus, the nutrient supply ratios vary around the Redfield ratios on a seasonal basis, with silicate and phosphorus in the shortest supply during the spring and nitrogen more likely to be limiting (based on ratios) during the rest of the year (Fig. 3).

Offshore nutrients, present and historical changes

There is a consistent westward and downstream transition away from the discharges of the Mississippi and Atchafalaya Rivers along the coastal plume in lower to higher salinities, higher to lower nutrients, and higher to lower surface chlorophyll *a* concentrations

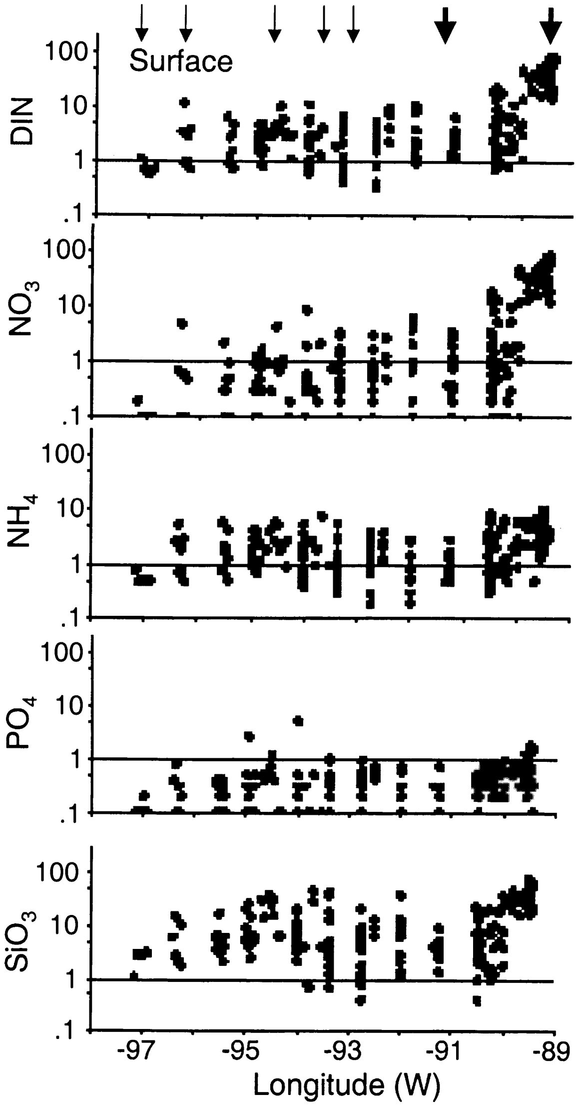

Figure 4. Concentration of dissolved inorganic nutrients (μM) in surface waters at stations with total depth between 10 and 100 m on the Louisiana continental shelf. The arrows indicate the source and relative contribution of freshwater inputs. Southwest Pass of the Mississippi River birdfoot delta is near 89.5° W and the Atchafalaya delta is near 91.5° W. (from Rabalais & Turner, 1998).

(Rabalais et al., 1996; Rabalais & Turner, 1998). The present distribution of dissolved inorganic nutrients is shown in Figure 4 for the area of the Louisiana shelf influenced by the Mississippi River system.

Probable nutrient limitation was assessed by Dortch & Whitledge (1992) and Justic et al. (1994) who compared the ambient nutrient concentrations with the k_S for nutrient uptake and, in the case of Si, a threshold value for uptake. Plots of relative frequencies (Justic et al., 1994) showed that dissolved N concentrations in the surface layer of the northern Gulf of Mexico during the period 1985–1992 were lower than 1 μM in about 13% of the cases. Reactive P was below 0.1 μM in 17% of the cases, while reactive Si concentrations lower than 2 μM occurred in 25% of the cases. These findings are important because studies of nutrient uptake kinetics (i.e. Rhee, 1973; Harrison et al., 1977; Goldman & Glibert, 1983; Nelson & Brzezinski, 1990) indicate that concentrations of 1 μM, 0.1 μM and 2 μM may be considered as threshold values for N, P and Si uptake, respectively. In contrast, the corresponding frequencies were estimated as 39%, 41% and 10%, respectively, in 1960, based on an historical reconstruction technique (Justic et al., 1995a). Thus, it appears that overall nitrogen and phosphorus nutrient limitation have decreased, while the probability of silicate limitation may have increased.

Fluctuations in the Si:N ratio within the riverine effluents and the offshore waters can affect diatom production and are believed to be major determinants in estuarine and coastal food web structure on a seasonal and annual basis, with major implications to oxygen and carbon cycling (Turner et al., 1998). Deviations from the 16:16:1 Si:N:P ratio in nutrients available in the water column may be a limiting factor for diatoms, as well as for other phytoplankton groups (Hecky & Kilham, 1988; Dortch & Whitledge, 1992). Also, a decreasing Si:N ratio may exacerbate eutrophication by reducing the potential for diatom growth in favor of noxious flagellates (Officer & Ryther, 1980). Another reasonable hypothesis that follows a more balanced nutrient composition, as evidenced in the Mississippi River and in the coastal waters as well, is that surface primary productivity has increased under these conditions.

Historical trends in productivity and hypoxia

One might expect a propensity for high productivity and development of hypoxia, given the high volume of fresh water and associated nutrients delivered by the Mississippi River into a stratified coastal system. Unfortunately, the long-term data sets that demonstrate changes in surface-water production and bottom water dissolved oxygen, such as available for the northern Adriatic Sea and areas of the Baltic and northwestern European coast, are few for the northern Gulf of Mexico. Therefore, biological, mineral or chemical indicators of surface-water production and hypoxia preserved in sediments that accumulate under the plume of the Mississippi River (in the area between transect C and the Mississippi River birdfoot delta, Fig. 1) must provide clues to prior hydrographic and biological conditions. Sediment cores analyzed for different constituents (Turner & Rabalais, 1994a; Eadie et al., 1994) document increased recent eutrophication and increased organic sedimentation in bottom waters, with the changes being more apparent in areas of chronic hypoxia and coincident with the increasing nitrogen loads from the Mississippi River system. Evidence comes from changes in silicate-based primary production, increased accumulation of diatom remains in the sediments, increased marine-origin carbon accumulation in the sediments, decreased diversity of selected benthic infauna, and relative changes in selected benthic infauna that indicate a worsening oxygen environment (Turner & Rabalais, 1994a, b; Eadie et al., 1994; Nelsen et al., 1994; Rabalais et al., 1996; Sen Gupta et al., 1996).

Human activities in the watershed undoubtedly changed the natural functioning of the Mississippi River system. Century-long patterns of freshwater discharge are not evident; thus, the long-term changes on the Louisiana shelf are linked primarily to the quality of the discharge (nutrient loads and ratios of nutrients) and not to the amount of fresh water. While century-long changes are evident in some of the retrospective analyses, the most dramatic and accelerating changes have been since the 1950s, when nitrogen loads began to increase, primarily from nitrate inputs, and eventually doubled to tripled over their historic values. The fact that the most dramatic changes in the continental shelf ecosystem have occurred since the 1950s and are coincident with an increase in nitrate flux, points to that aspect of human ecology for future management scenarios.

Dilution, uptake and regeneration of nutrients

Despite the extremely high nutrient inputs to the shelf, nutrient concentrations are depleted to low, and sometimes undetectable, levels within a short distance of the river mouth (Lohrenz et al., 1990, 1997, 1999a; Rabalais et al., 1991, 1996, 1998; Dortch & Whitledge, 1992; Nelson & Dortch, 1996; Rabalais & Turner, 1998; see Fig. 4). Nitrate, the major form of nitrogen supplied by the river, decreases quickly. Consequently ammonium, resupplied by regeneration (Bode & Dortch, 1996), is the only available nitrogen source over much of the shelf, but it is present at much lower concentrations than nitrate is initially. Dissolved organic nitrogen (DON) is supplied by the river, but its dynamics in the outflow of the Mississippi River are complex, and it is unclear if it is a source or a sink for nitrogen available to phytoplankton (Lopez-Veneroni & Cifuentes, 1994). Silicate can be depleted to extremely low levels. In fact some of the lowest concentrations of silicate observed in any ocean were measured on the Louisiana shelf (Nelson & Dortch, 1996; Dortch, unpubl. data). Dissolved inorganic phosphate concentrations also decrease to low levels, but are usually detectable. The importance of dissolved organic phosphorus has not been fully assessed, although it may be recycled rapidly (Ammerman, 1992; Ammerman et al., 1995). The area with high nutrient concentrations varies, depending on river flow and season, but is much smaller than the total area of the Louisiana/Texas shelf impacted by the Mississippi River. High phytoplankton biomass and productivity can be maintained due to high rates of nitrogen and phosphorus regeneration (Ammerman, 1992; Gardner et al., 1994; Ammerman et al., 1995; Bode & Dortch, 1996). Short-term rates of Si regeneration are generally much lower, which increases the potential for Si limitation (Nelson & Dortch, 1996).

The rapid depletion of nutrients is due to biological uptake and conservative mixing between high nutrient Mississippi river water and low nutrient, high salinity Gulf of Mexico water, with the relative importance of these processes depending on the season and river flow. At most times, there are large negative deviations from the conservative mixing line in estuarine mixing diagrams of nutrient concentration versus salinity, that are indicative of high biological removal (Dortch & Whitledge, 1992; Turner & Rabalais, 1994b; Bode & Dortch, 1996; Nelson & Dortch, 1996; Lohrenz et al., 1999a). Linear or non-conservative mixing diagrams (Lohrenz et al., 1999) suggest other sources of nutrients, such as low salinity nutrient sources (desorption from sediments or production of dissolved organic nitrogen) or intrusion of deeper offshore nutrients (Fox et al., 1985; Lopez-Veneroni & Cifuentes, 1994). Although the N:P ratio in river water is near the Redfield ratio and suggests that either may be limiting primary production in receiving Gulf waters, the efficient remineralization of P in marine systems coupled with the inevitable losses of N due to denitrification probably means that N is the limiting nutrient (of N and P) beyond the immediate plume and may explain some of the nonlinear mixing diagrams in Fox et al. (1985), Hitchcock et al. (1997) and Lohrenz et al. (1999a). 'New' nutrients become depleted along the river-to-ocean mixing gradient through dilution and biological uptake, and regenerated water column nutrients support primary production for great distances from the river mouth (Bode & Dortch, 1996; Nelson & Dortch, 1996). Additional nutrients become available through regenerative processes in the sediments, but their resupply to the upper mixed layer is diminished by the presence of a strong pycnocline that can be disrupted by wind-mixing events.

Nutrient-enhanced productivity

High biological productivity in the immediate (320 g C m^2 y^{-1}) and extended plume (290 g C m^2 y^{-1}) of the Mississippi River (Lohrenz et al., 1990; Sklar & Turner, 1981; respectively) is mediated by high nutrient inputs and regeneration, and favorable light conditions. Small-scale and short-term variability in productivity are the consequence of various factors, such as nutrient concentrations, temperature, salinity, light and mixing rates (Lohrenz et al., 1990, 1994), but are also clearly influenced by Mississippi River flow and nutrient flux to the system (Justic et al., 1993, 1997; Redalje et al., 1994; Lohrenz et al., 1997).

Lohrenz et al. (1997) clearly demonstrated that primary production in shelf waters near the delta and to some distance from it was significantly correlated with nitrate and nitrite concentrations and fluxes over the period 1988–1994 (Fig. 5). Light limitation was likely an important factor during winter months, but a positive correlation was demonstrated between river inputs of nitrate and nitrite for other times of the year. The relationships between riverine flux and concentration for those stations on the western end of their study area (i.e. near transect C in Fig. 1) were improved when the riverine input data were lagged one month. These results are consistent with those of Justic et al. (1993, 1997) for a one-month lag between net production in surface waters and river discharge and nitrate flux. Even stronger correlations were observed by Lohrenz et al. (1997) between the concentration of orthophosphate and primary production, but these were not significant (smaller sample size).

Variability in primary production in the region of the northern Gulf of Mexico impacted by the Mississippi River is quite high, due to the dynamic and heterogeneous conditions found in the river/ocean mixing zone. Highest primary production is typically observed near the river delta (Lohrenz et al., 1999a, b). Maximum values of biomass and primary production were typically observed at intermediate salinities within the Mississippi River plume and coincided with non-conservative decreases in nutrients along the salinity gradient (Lohrenz et al., 1999a). Rates of primary production along a salinity gradient of the Mississippi River plume were constrained by low irradiance and mixing in the more turbid, low salinity regions of the plume, and by nutrient limitation outside the plume. This pattern of localized maxima in phytoplankton biomass and production attributable to declining turbidity in the presence of relatively high nutrient levels has been reported for other major rivers including the Amazon (DeMaster et al., 1986; Smith & DeMaster, 1996), Huanghe (Turner et al., 1990) and Changjiang (Xiuren et al., 1988). Similar relationships of chlorophyll *a* biomass with salinity were observed across the broad region of the Louisiana shelf influenced by Mississippi River discharge (Fig. 6). A gradient of decreasing chlorophyll *a* biomass was observed across the Louisiana shelf in a westerly direction from the Mississippi River delta (Fig. 7).

There was a high degree of coherence between Mississippi River nitrate fluxes and net production rates at station C6* (Fig. 1) for 1985–1992 (Fig. 8) (Justic et al., 1997). Mississippi River discharge for 1985–1992 had a typical sinusoidal seasonal pattern consistent with longer records (Bratkovich et al., 1994) with highest nitrate flux in April and lowest in September. Although the monthly cycle of Mississippi River nitrate flux generally resembled the monthly cycle of freshwater runoff, the peak in nitrate flux was somewhat delayed with respect to the peak in freshwater runoff as a result of the seasonal pattern in riverine nitrate concentrations. The net production rates for the upper water column (Justic et al., 1997) showed a well-defined seasonal cycle with a minimum of −0.2 g C m^2d^{-1} in December and a maximum of 1.2 g C m^2 d^{-1} in April. A cross correlation function of net production and riverine nitrate flux showed that the two series were highly correlated ($CCF = 0.73$, $p < 0.01$) and that a seasonal maximum in net production lags with respect to a riverine nitrate maximum by one month (Fig. 8).

The downstream transition away from the discharges of the Mississippi and Atchafalaya Rivers in lower to higher salinities, higher to lower nutrients, and higher to lower surface chlorophyll *a* concentrations (Figs 4 and 7) are further reflected in the flux of

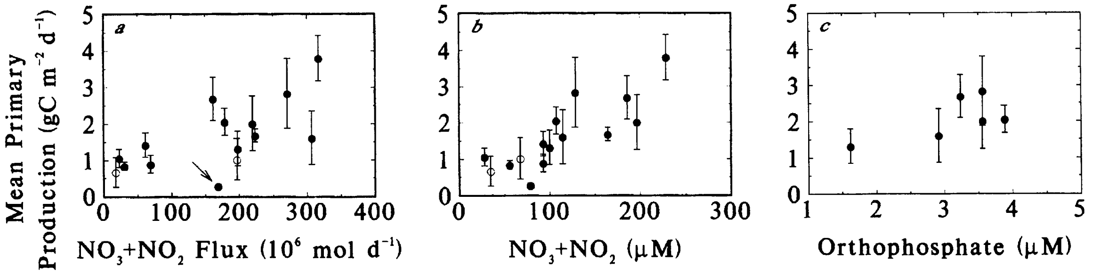

Figure 5. Relationship between mean primary production for the combined central and eastern regions of the Mississippi River bight and (a) riverborne nitrate and nitrite flux, (b) nitrate and nitrite concentration at Venice and (c) orthophosphate concentrations at Belle Chase. Historical primary production data (o) from Thomas and Simmons (1960) are included in (a) and (b) for comparison. Error bars are ± 1 s.e. (from Lohrenz et al., 1997; used with permission of Inter-Science Research Publisher).

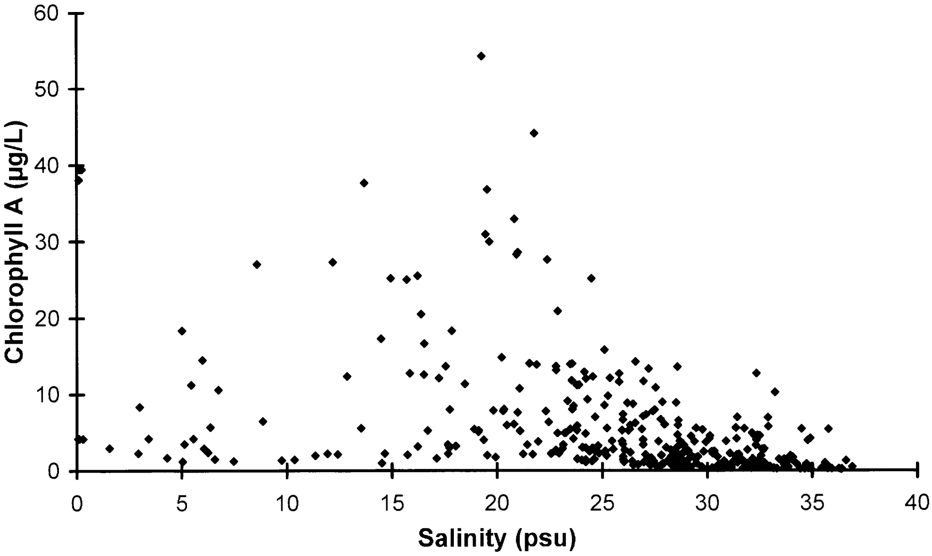

Figure 6. Relationship of surface chlorophyll *a* and salinity for six cruises (April 1992, October 1992, April 1993, July 1993, April 1994, July 1994) from 89.5° W to 97° W in water depths of 10–100 m. Maximum value of 209 μg l^{-1} was deleted from the plot (from Rabalais & Turner, 1998).

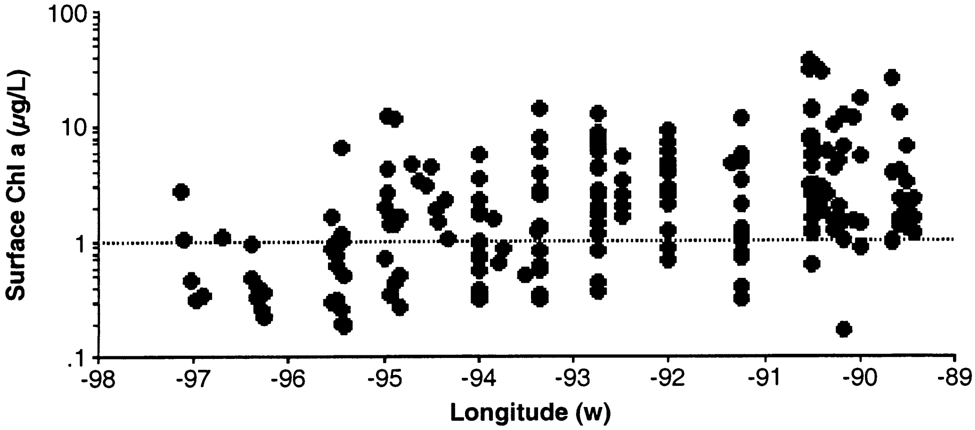

Figure 7. The concentration of chlorophyll *a* in surface waters at stations between 89.5° W and 97° W and water depths of 10 and 100 m for the periods April 1992, October 1992, April 1993, July 1993, April 1994, July 1994 (redrawn from Rabalais & Turner, 1998). Maximum value of 209 μg l^{-1} was deleted from the plot. Southwest Pass of the Mississippi River birdfoot delta is near 89.5° W and the Atchafalaya delta is near 91.5° W.

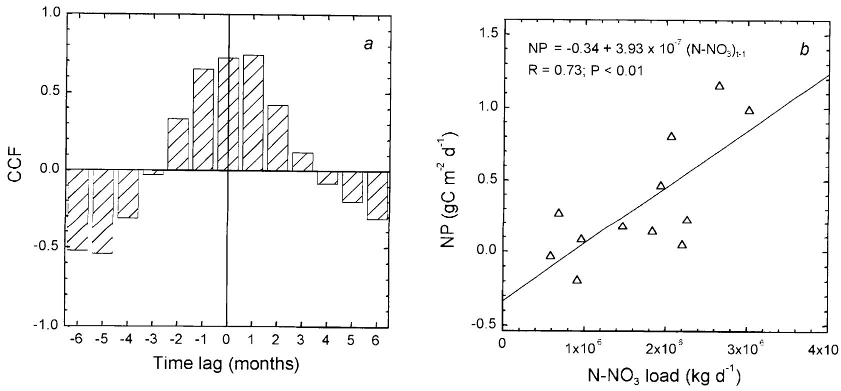

Figure 8. Left panel: Cross correlation function (CCF) for Mississippi River nitrate flux at Tarbert Landing and net production of the upper water column (1–10 m at station C6* in 20-m depth off Terrebonne Bay, see Fig. 1). Right panel: best-fit time-delayed linear model for the regression of net production (NP) on nitrate load. The model is $NP_t = -0.34 + 3.93 \times 10^{-7}$ $nitrate_{t-1}$ where t and t-1 denote values for the current and preceding months, respectively. Symbols denote monthly averages for the period 1985–1992 (from Justic et al., 1997; used with permission of Inter-Science Research Publisher).

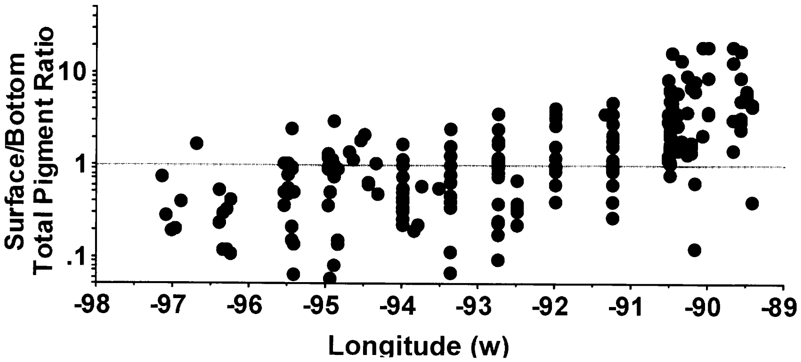

Figure 9. Relationship between the ratio of the concentration of total phytoplankton pigments ($\mu g\ l^{-1}$) in surface to bottom waters with longitude at the sampling stations between 89.5° W and 97° W and water depths of 10 and 100 m for the periods April 1992, October 1992, April 1993, July 1993, April 1994, July 1994 (from Rabalais & Turner, 1998). Southwest Pass of the Mississippi River birdfoot delta is near 89.5° W and the Atchafalaya delta is near 91.5° W.

organic material as seen in surface-to-bottom pigment ratios (Fig. 9) and accumulation of phaeopigments in the lower water column. Respiration rates are higher in shallower waters and are related to chlorophyll *a* concentrations (Turner & Allen, 1982; Turner et al., 1998; Turner & Rabalais, 1998). The distribution of low dissolved oxygen in bottom waters is related to high surface net production, but this relationship is lagged in time and (Justic et al., 1993; Rabalais et al., 1994). Phytoplankton and fecal pellets in surface waters fall into the bottom layers quickly, perhaps in a few days or less, but surface and bottom currents are not traveling in the same direction or at the same speed. Most of the organic matter reaching the bottom is not consumed in hours, but many days (and probably weeks). The depletion of oxygen is cumulative, and depends also on the reaeration rate (Justic et al., 1996). No consistent patterns should therefore be expected between phytoplankton pigments in surface waters and low oxygen in bottom waters for a specific location in time. There is, however, a consistent transition away from the river discharges along the coastal plume in flux of organic material, respiration rates and incidence of bottom water hypoxia (qualitatively demonstrated in Fig. 1, frequency distribution of mid-summer hypoxia down-current from the Mississippi and Atchafalaya River plumes).

Phytoplankton in the region of station C6* (Fig. 1) can be divided into three groups, picocyanobacteria, diatoms, and others (mostly small flagellates) (Fig. 10), although dinoflagellate blooms do occur sporadically (Dortch, 1998; Dortch, unpubl. data).

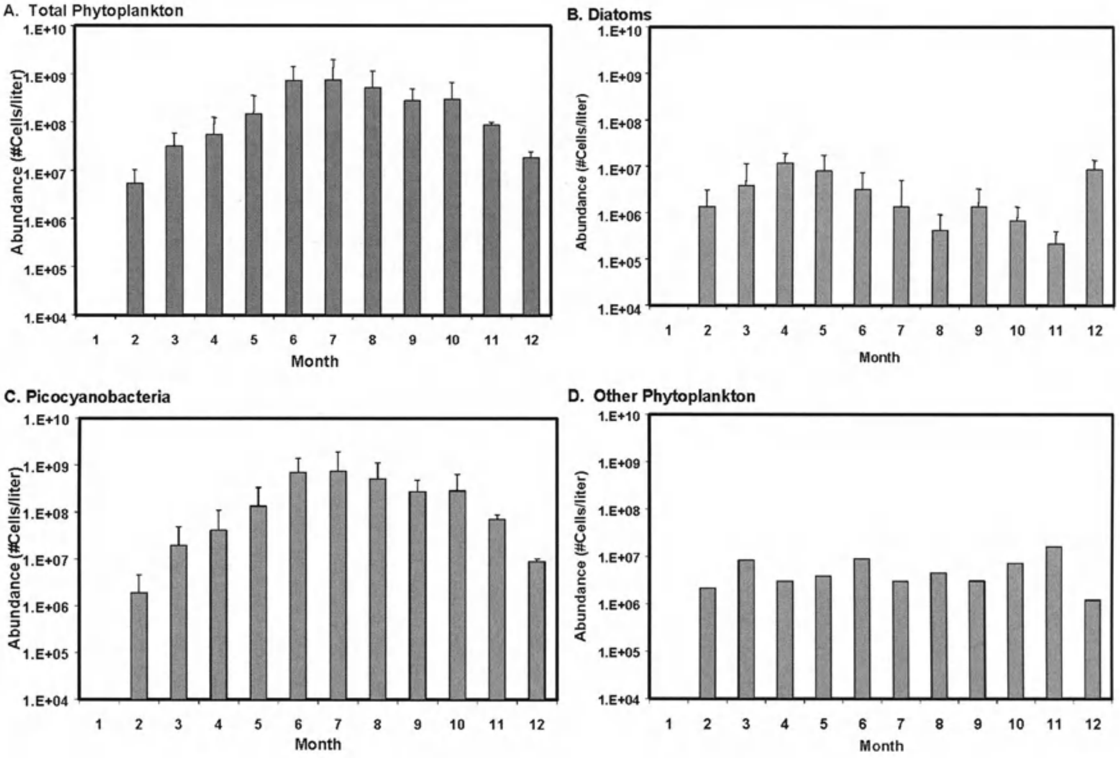

Figure 10. Average monthly surface layer phytoplankton major taxa composition at station C6A and C6B (see Fig. 1) from 1990 to 1995 (mean ± 1 S.E.) (Dortch, unpubl. data). Number of samples ranges from 14 to 41 for all months except February ($n = 5$) and December ($n = 2$).

Although the small flagellates can be numerically abundant, they are much less abundant than picocyanobacteria, much smaller than most diatoms and unlikely to dominate the biomass. Further, there is relatively little seasonal variation in flagellate abundance ('other' in Fig. 10). Thus, picocyanobacteria and diatoms are the most important phytoplankton groups in the region influenced by the Mississippi River. Picocyanobacteria are most abundant in the summer and early fall, whereas diatoms are most abundant in the spring. In terms of estimated biomass, diatom biomass usually dominates, especially in the spring. The exception was during the Mississippi River flood in July 1993, when all phytoplankton were elevated, but especially picocyanobacteria (Dortch, 1994; Rabalais et al., 1998).

Nutrient limitation of phytoplankton growth

Despite the extremely high nutrient inputs to the shelf, nutrients are depleted to low, and sometimes undetectable, concentrations within a short distance of the river mouth (Lohrenz et al., 1990, 1997, 1999a; Dortch & Whitledge, 1992; Bode & Dortch, 1996; Nelson & Dortch, 1996; Rabalais et al., 1996, 1998). Patterns of nutrient depletion provide evidence that riverine inputs of nutrients and their pattern of regeneration ultimately limit the extent of river-enhanced production and biomass. Ambient nutrient concentrations and ratios, bioassay experiments, and other indicators of nutrient limitation (Rabalais et al., 1999) all suggest that N, P or Si may be limiting at some times and places in the outflow of the Mississippi River. The details of when and where particular nutrients are limiting and the severity of limitation are not completely known. N limitation occurs most often at higher salinities and during low flow periods. In contrast, P limitation occurs mostly at intermediate salinities and during high freshwater input. Si limitation, which affects diatom growth, appears to be more spatially and temporally variable than P or N limitation but is more prevalent in spring than in summer. Phytoplankton species may be differentially susceptible to nutrient limitation, with an obvious example being the requirement of diatoms for Si, and there are species differences among diatoms with regard to Si limitation. Although limitation by both Si and P does occur, N limitation is more frequent and extends over a larger area. Consequently, the rate of nitrogen loading is believed to be a critical factor in regulating the overall production of phytoplankton over the broad region influenced by the river and affected by bottom-water hypoxia.

Fate of carbon

The spring delivery of nutrients initiates a seasonal progression of biological processes that ultimately leads to the depletion of oxygen in the bottom waters. One might expect that the vertical export of particulate organic carbon (POC) would also be high, given the high rates of primary production, and be roughly proportional to the quantity of carbon fixed in the surface waters (Suess, 1980). Although the overall flux of POC on the continental shelf influenced by the Mississippi River is high, the relationship between POC export and primary production is quite variable in time and space. Redalje et al. (1994) examined the relationship between primary production and the export of POC from the euphotic zone determined with free-floating sediment traps. Productivity and POC exports exhibited similar trends in spring and fall, but were uncoupled in summer. The lowest ratio of export to production coincided with the time when production was greatest, and the highest ratios occurred when production was the lowest. Export ranged from low values of 3–9% during July–August 1990 to high values during March 1991, when export exceeded measured water column-integrated primary production by a factor of two.

In another study of the vertical flux of particulate material, particle traps were deployed on an instrument mooring within the zone of recurring hypoxia at depths of 5 m and 15 m in a 20-m water column in spring, summer and fall of 1991 and 1992 (station C6*, Fig. 1) (Qureshi, 1995). Carbon flux was approximately 500–600 mg C m^{-2} d^{-1} in 15-m water depth (Qureshi, 1995). A rough estimate of the fraction of production exported from the surface waters (compared to seasonal primary production data of Sklar & Turner, 1981) was highly variable and ranged from 10 to 200% with higher percentages in spring (Qureshi, 1995). A large proportion of the POC that reached the bottom was incorporated in zooplankton fecal pellets (55%), but also as individual cells or in aggregates. Both phytoplankton and zooplankton fecal carbon flux were greater in the spring and the fall than in the summer.

The high particulate organic carbon flux to the 15-m moored trap was sufficient to fuel hypoxia in the bottom waters below the seasonal pycnocline (Qureshi, 1995; Justic et al., 1996). The flux of organic material in summer, while it sustained hypoxia, was incremental to the majority flux of particulates in the spring (Qureshi, 1995). The moored sediment traps were not deployed from late fall through early spring, and fluxes during that period are not known.

The oxygen consumption rates in near-bottom waters of the seasonally oxygen-deficient continental shelf were measured during spring and summer cruises in several years (Turner & Rabalais, 1998; Turner et al., 1998). Respiration rates varied between 0.0008 and 0.29 mg O_2 l^{-1} h^{-1}, and were sufficient to reduce the *in situ* oxygen concentration to zero in less than four weeks. The rates were inversely related to depth and decreased westward of the Mississippi River delta, consistent with the decrease in nutrients, chlorophyll *a* and total pigment concentrations, and the relative proportion of surface-to-bottom pigments. The amount of phytoplankton biomass in the bottom waters across the Louisiana inner and middle continental shelf is high, often exceeding 30 μg l^{-1}, and a high percentage is composed of phaeopigments (Rabalais & Turner, 1998). The respiration rate is proportional to phytoplankton pigment concentration (Turner & Allen, 1982), and, thus, higher rates of oxygen consumption would be expected where higher flux of material reaches the lower water column and sediments. Respiration rates per unit phytoplankton pigment were highest in the spring, in shallower waters, and also closest to the Mississippi River delta.

Composition of sinking material and nutrient availability

On the Louisiana shelf in the area influenced by the Mississippi River, the prevalence of Si limitation, indicated by nutrient concentrations and ratios, suggests that diatoms may at times be Si limited, whereas non-diatoms are not (Dortch et al., 2001; Dortch, unpubl. data). Several lines of evidence support this contention. (1) The annual average abundance of diatoms is inversely proportional to the prevalence of Si limitation in the same year, except in 1992, which was unusual for other reasons. (2) Cell count bioassay experiments show that diatoms are limited by Si, N or Si + N availability, sometimes when other phytoplankton are limited by another factor. (3) A shortage of silicate does not just modify the relative abundance of diatoms and non-diatoms. It also influences the diatom species composition so that when Si is limiting, diatoms with low Si requirements, as manifested by light silicification of the frustule (Dortch et al., 1992; Fahnenstiel et al., 1995) or lower k_S for silicate uptake (Nelson & Dortch, 1996), predominate.

Despite the limited carbon flux data, there were some clear links between different sources of carbon flux and nutrient availability in the coastal zone (Dortch et al., 1992, 2001; Fahnenstiel et al., 1995). Because of its effect on diatom abundance, silicate availability may be a factor controlling phytoplankton and fecal pellet carbon flux. Diatoms comprised a much higher percentage of the total phytoplankton sinking into the moored sediment traps (Qureshi, 1995) than they were in the surface water (Dortch, unpubl. data), suggesting selective sinking of diatoms. The other phytoplankton found in sediment traps were primarily picocyanobacteria that did not contribute substantially to the carbon flux because of their small size. Because the peak in cyanobacterial flux was greater in spring when diatom abundance was greatest than it was in summer and fall when cyanobacterial abundances were highest, it has been proposed that the cyanobacteria sink as part of diatom aggregates (Dortch et al., 1997, 2001). The diatom species that did sink represent only part of the total diatom community, including heavily to moderately silicified species and excluding lightly silicified species (Dortch et al., 1992, 1997, 2001; Fahnenstiel et al., 1995). Thus, Si must be available for high sinking flux of heavily to moderately silicified diatoms. There is an apparent contradiction in these results. The greatest sinking fluxes occur when diatom abundances are high, which is also when Si limitation is much more likely to occur. Other data suggest that Si limitation triggers sinking in many diatom species (e.g. Bienfang et al., 1982).

Many zooplankton feed on diatoms, which are the most abundant food in the larger size range in this system. Fecal pellet production is often proportional to the availability of food (e.g. Corner et al., 1972; Butler & Dam, 1994). In 1991 there was a close correspondence between fecal pellet flux and diatom abundance, although in 1992 there was not (Rabalais et al., 1999; Dortch et al., 2001). With carbon flux data for only two years (Qureshi, 1995), one year cannot be labeled as unusual, but other data suggest that 1992 may have been unusual. Because diatom abundance is dependent on silicate availability (Dortch, unpubl. data; Rabalais et al., 1999), this implies that Si availability may be a factor controlling fecal pellet production and flux.

Turner et al. (1998) combined the sediment trap fecal pellet carbon data (Qureshi, 1995), chlorophyll data, zooplankton data and seasonal and depth-related respiration rate experiments in an analysis of how these relationships varied around a Si:DIN ratio of 1:1 as delivered by the Mississippi River (lagged three months). They showed that (1) there is a strong vertical, rather than horizontal, coupling between oxygen consumption in bottom waters and organic loading from surface waters, and (2) higher water-column respiration rates are driven by river-derived nutrients stimulating *in situ* organic production that sinks to the bottom layers. They also showed that respiration rates in bottom waters were responsive to zooplankton fecal pellet production (as predicted by Qureshi, 1995) and to diatom production. The trophodynamics of carbon production, zooplankton, carbon flux and respiration are sensitive to the Si:DIN ratio in river waters.

Oxygen and carbon budgets

Oxygen profiles and calculations of oxygen anomalies for station C6* (Fig. 1) over the period 1985–1992 were used to define oxygen and carbon budgets for an area that is consistently hypoxic on a seasonal basis (Justic et al., 1996, 1997). This area is suitable for the development of a coupled biological–physical two-box model for several reasons. Vertical oxygen transport is likely to be more important than horizontal oxygen transport for this area, because the data suggest a relatively high coherence between changes in vertical temperature gradients and changes in bottom oxygen concentration (Rabalais et al., 1992) and a strong tidal signal of any kind which would indicate horizontal transport is not present in the periodograms of oxygen data from station C6* (Rabalais et al., 1994). The surface-water layer, above the prevalent pycnocline at 10 m, shows an oxygen surplus during February-July with the maximum in April and May that coincides with mean peak Mississippi River flow. An oxygen surplus also means that there is an excess of organic matter derived from primary production that can be redistributed within the system; much of this will eventually reach the lower water column and sediments. The bottom layer, below the pycnocline to

20 m, exhibits an oxygen deficit throughout the year, but reaches its highest value in July when surface-to-bottom density differences are greatest. Bottom hypoxia in the northern Gulf is most pronounced during periods of high water column stability when surface-to-bottom density differences are greatest (Rabalais et al., 1991; Wiseman et al., 1997). The correlation between Mississippi River flow and surface oxygen surplus peaks at a time-lag of one month, and the highest correlation for bottom oxygen deficit is for the time-lag of two months (Justic et al., 1993). These findings suggest that the oxygen surplus in the surface layer following high flow depends on nutrients ultimately coming from the river but regenerated many times.

About 90% of the annual net oxygen production at station C6* occurs between February and June (Justic et al., 1996, 1997). The integrated annual net productivity (NP) of the upper water column (0–10 m) at station C6* is 423 g O_2 m^{-2} (Fig. 11). If an oxygen-to-carbon ratio of 3.47 by weight (mol O_2:mol C = 138:106; PQ = 1.3) is assumed for the photosynthetic process, then the total net carbon production is 122 g C m^2 y^{-1}. The excess of organic matter, derived from primary production, is redistributed within the system, and eventually decomposed in the lower water column and in the sediments. Integrated oxygen uptake rates for the lower water column (10–20 m) are significantly higher between January and June than the rest of the year. This correlates well with the seasonal changes in NP in the upper water column. The integrated annual oxygen uptake rate in the lower water column at station C6* is 197 g O_2 m^{-2} y^{-1}, which converts to a value of 57 g C m^{-2} y^{-1}, if an RQ value of 0.77 (mol C:mol O_2 = 106:138) for the respiration process. Thus, on an annual basis, 47% of the surface net organic production at station C6* is decomposed in the lower water column and in the sediments (TR:NP = 0.47), a value that compares well with estimates of fixed carbon exported from surface waters to the lower water column (Qureshi, 1995).

A high degree of coherence exists between the Mississippi River nitrate flux and net production rates at station C6*. This allowed Justic et al. (1997) to predict that the net productivity of the upper water column has increased since the 1950s coincident with increased river nitrate flux and concentration – a relationship verified with sediment cores. If one assumes that the riverine nitrogen input was 50% lower than at present, the monthly nitrate flux during the 1950s did not exceed 1.6×10^6 kg d^{-1}. Based on the relationships of the model (Justic et al., 1996, 1997), that flux would be sufficient to support net productivity of about 0.29 g C m^{-2} d^{-1}, which is only 25% of the peak NP value for 1985–1992 (1.15 g C m^{-2} d^{-1}). Consequently, the integrated annual net production was substantially lower than at present, and probably did not exceed 25 g C m^{-2} y^{-1}. Even if losses were due to export, burial were not significant, and the total annual production of 35 g C m^{-2} $y^{-1}$was decomposed in the lower water column and sediments, the annual total oxygen uptake value (TR) would have been around 121 g O_2 m^{-2} y^{-1}. This result is substantially lower when compared with the estimates for 1985-1992 (197 g O_2 m^{-2} y^{-1}) and 1993 (195 g O_2 m^{-2} y^{-1}). Thus, the net productivity of the upper water column appears to be an important factor controlling the accumulation of organic matter in sediments and development of hypoxia in the lower water column. The model for station C6*, calibration data, experimental results from the 1993 flood, and doubled CO_2 climate scenario projections (20% increase in river discharge in May and June), as well as modifications to coastal carbon and oxygen budgets under anthropogenic nutrient enrichment throughout the world, indicate a close coupling between river-borne nutrients, net productivity, vertical carbon flux, and hypoxia on decadal time scales.

Summary of evidence for nitrogen-stimulated productivity

A series of experiments, empirical relationships, seasonal oxygen and carbon budgets, time series analyses, models and comparisons with other regions of the world's coastal ocean indicate a close coupling between river-borne nutrients, net productivity, vertical carbon flux and hypoxia on short (day) to long (decadal to century) time scales (Table 1).

Models that link nutrients, productivity and hypoxia

There are clearly demonstrated empirical relationships between primary production and dissolved inorganic nitrogen concentration and flux and orthophosphate concentration (Lohrenz et al., 1997), and of net productivity with discharge and nitrate loads (Justic et al., 1993, 1997). Similar relationships exist between Atchafalaya River discharge and oxygen depletion on

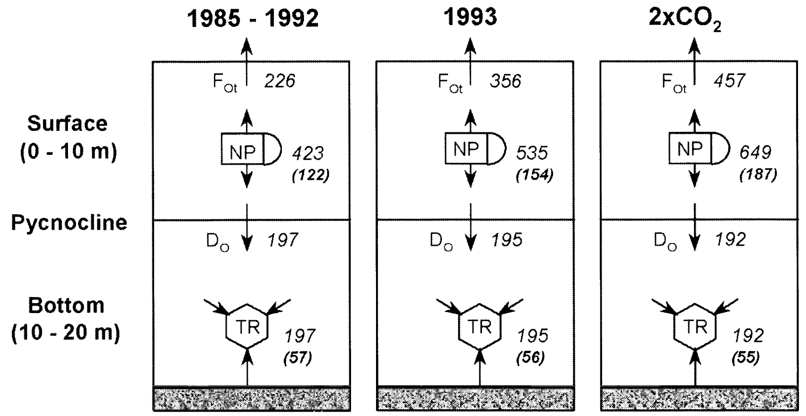

Figure 11. Global oxygen fluxes (g O_2 m^{-2} d^{-1}) at station C6* for the periods 1985–1992 and 1993 and model projections for a doubled CO_2 climate. F_{Ot} denotes the total air-sea oxygen flux, NP is the net productivity of the upper water column (0–10 m), D_O is the diffusive oxygen flux through the pycnocline, and TR is the total oxygen uptake in the lower water column (10–20 m). Carbon equivalents, computed from the Redfield stoichiometric model C:O_2 = 0.288, by weight), are given in parentheses (from Justic et al., 1997; used with permission of Inter-Science Research Publisher).

Table 1. Evidence for nitrogen driven phytoplankton production (modified from Rabalais et al., 1999)

Temporal scale	Evidence
Days	Bioassay experiments.
	Simulated *in situ* measurements of primary production across a range of dissolved inorganic nitrogen concentrations.
	Short-term primary production models.
	Correlation of nitrate with primary production.
	Depletion of nitrate and silicate along a salinity dilution gradient.
	in situ Redfield ratios.
Months	Correlation of primary production with time-lagged nutrient concentration and flux.
	Correlation of surface-water net production with time-lagged nutrient flux.
	Correlation of surface-water net production and bottom water oxygen stress with 1-mo and 2-mo, respectively, lagged freshwater discharge.
	Oxygen and carbon budgets.
	Carbon flux relationships with indicators of river discharge and surface-water increased production.
	Response of mass balance model to reductions in nitrogen load (see below).
Years	Sediment cores and coincidental timing with increased nitrogen loading.
	Increase in accumulation of marine-source carbon.
	Increase in silicate-based productivity.
	Increase in foraminiferal index of A/E (increased carbon accumulation and worsening oxygen stress).
	1998, 1992, or other low discharge years for non-events; variability in spring discharge, predicted C flux, or stratification, or combination.

the southwestern Louisiana shelf (Pokryfki & Randall, 1987). Justic et al. (1996, 1997) further explored carbon and oxygen budgets at a station within the core of the hypoxic zone with a physical–biological coupled two box model (Fig. 11).

Another mathematical model was developed for the Louisiana inner shelf influenced by the discharge of the Mississippi and Atchafalaya Rivers that synthesized environmental data over the broad area of influence (as opposed to the Justic et al. model for station C6* on the southeastern shelf) in order to provide an understanding of environmental processes controlling primary productivity and bottom-water dissolved oxygen (Bierman et al., 1994, 1999). This model (hereafter called the NECOP model [Nutrient Enhanced Coastal Ocean Productivity]) was also used to assess how biological processes might be influenced by reductions in nutrient loads from the Mississippi River system (Limno-Tech, 1995; Bierman et al., 1999; Brezonik et al., 1999).

Bierman et al. (1994) applied a version of the U.S. Environmental Protection Agency WASP (Water Quality Analysis Simulation Program; Ambrose et al., 1993) to the Louisiana inner shelf region affected by hypoxia. The conceptual framework for this model is shown in Figure 12. State variables in the model include salinity, phytoplankton carbon, phosphorus (dissolved orthophosphate and organic forms), nitrogen (ammonium, nitrate plus nitrite and organic forms), dissolved oxygen and carbonaceous biochemical oxygen demand (CBOD). User-specified external forcing functions include constituent mass loadings, advective-dispersive transport, seaward boundary conditions, sediment fluxes, water temperature, incident solar radiation and underwater light attenuation. Sediment interactions are represented by user-specified values for net settling rates for particulate phase constituents, sediment–water diffusive fluxes for dissolved nutrients and sediment oxygen demand (SOD).

The spatial domain of the model is represented by a 21-segment water column grid extending from the Mississippi River delta west to the Louisiana-Texas boarder, and from the shoreline seaward to the 30–60-m isobath (Fig. 13). There is one vertical mixed layer nearshore (average depth of 5.6 m) and two vertical mixed layers offshore separated at an assumed pycnocline depth of 10 m. The bottom offshore segments range in thickness from 6.1 to 20.3 m below the pycnocline.

The segmentation for the NECOP model was determined originally by the distribution of historical water quality data (primarily the hypoxia studies of Rabalais, Turner & Wiseman). Salinity was used to identify characteristic water masses and to determine the geometric boundaries of the grid. The coarse scale of the model is limited in two principal ways: (1) near-field horizontal gradients in the vicinity of the Mississippi and Atchafalaya River plumes are not explicitly represented, and (2) important vertical scale characteristics are not fully represented. Rabalais et al. (1994, 1996, 1999) have shown that the bottom-water oxygen mass changes configuration in response to winds, currents and tidal advection and that hypoxia can occur not only at the bottom near the sediments, but also well up into the water column.

The temporal domain of the model represents steady-state, summer average conditions, and thus represents only a single 'snapshot' in time, as opposed to the Justic et al. (1996) model that changes by month but represents only a single station. In reality, there is great daily and weekly variability in current flow and stratification on the Louisiana inner shelf (Rabalais et al., 1996), but the NECOP model was constrained by the availability of a single shelfwide monitoring cruise each year during July–August to delimit the area of hypoxia. The NECOP model was calibrated to a comprehensive set of field data collected during July 1990 at over 200 stations in the northern Gulf of Mexico (Bierman et al., 1994). Reasonable comparisons were obtained between computed and observed values for model state variables, primary productivity and mass settling fluxes for particulate carbon and nitrogen. The model calibration was extended to include earlier historical data collected during 1985–1988 (Rabalais et al., 1999). The calibrated model was used for diagnostic analyses, sensitivity analyses and numerical experiments. Details of the model calibration are available in Bierman et al. (1994) and Limno-Tech (1995).

Forecast simulations of nutrient load reductions

In the following paragraphs, we present results of simulations designed to test whether water quality parameters on the Louisiana inner shelf are responsive to changes in nutrient loads from the Mississippi River system. Year-specific forcing functions were applied to 1985, 1988 and 1990 summer average conditions (Table 2). These forcing functions represent average Mississippi and Atchafalaya River conditions for antecedent periods of approximately one month. Inflow to the model grid from the Mississippi River was much lower than inflow from the Atchafalaya River during the July 1990 application in order to match observed salinities in the model segments near the delta that indicated a net eastward drift in water circulation during that period.

The calibrated NECOP water quality model was run for simulations that reduced N and P loads from the Mississippi and Atchafalaya River system by 10–70%. To address uncertainties due to potential differ-

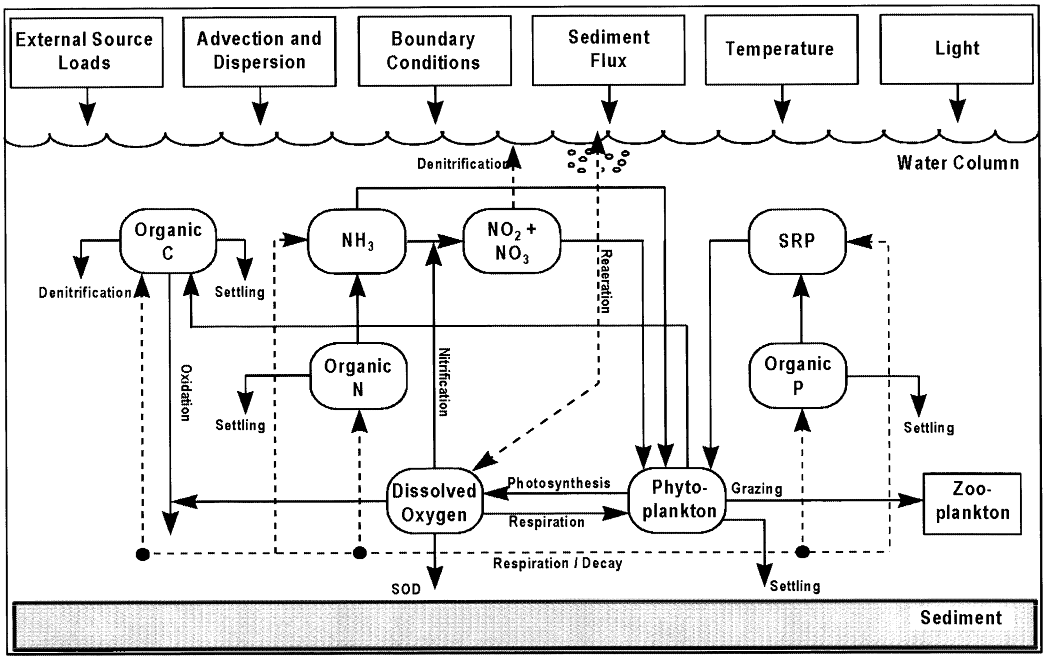

Figure 12. Schematic diagram of principal model state variables and processes in the NECOP water quality model (from Bierman et al., 1994; used with permission of the Estuarine Research Federation).

Table 2. Tributary inflows and nutrient loads in base calibration

Parameter	Mississippi			Atchafalaya		
	1985	1988	1990	1985	1988	1990
Inflow[a] (m^3 s^{-1})	4515	1760	1100	3829	1399	5700
N Load (metric tons d^{-1})	956.0	149.4	294.0	595.0	114.8	911.0
Inorganic[b]	671.0	116.0	230.0	324.0	62.8	458.0
Organic	285.0	33.4	64.2	271.0	52.0	453.0
P Load (metric tons d^{-1})	78.0	15.2	28.5	33.0	9.1	98.6
Available[c]	46.8	9.1	5.7	19.8	3.6	29.6
Unavailable[d]	31.2	6.1	22.8	13.2	5.4	69.0

[a] Sum of Mississippi inflows from Southwest Pass and westward flows from other passages.
[b] Sum of ammonium and nitrate plus nitrite.
[c] Dissolved orthophosphorus.
[d] Total phosphorus minus dissolved orthophosphorus.

ences in environmental conditions, separate forecast simulations were conducted for July 1985, August 1988 and July 1990 for each load reduction. To address uncertainties in specification of external boundary conditions, each load reduction simulation was conducted under two assumptions: (1) all seaward and sediment boundaries held constant at base-calibration values, and (2) all boundary conditions reduced by the same percentage as the imposed nutrient loading of the simulation. These assumptions were necessary because (1) the boundary conditions are not computed by the model, but must be externally specified using available field data, and (2) values for the boundary conditions are not directly dependent on the river nutrient loads, but can be expected to decrease as the nutrient loads decrease. This approach brackets results of the simulations between present conditions and estimates of future conditions and was necessary because of the model's limited spatial domain; the seaward boundaries are not far enough removed to be independent of sources of nutrients. The necessity to bracket results between different sediment

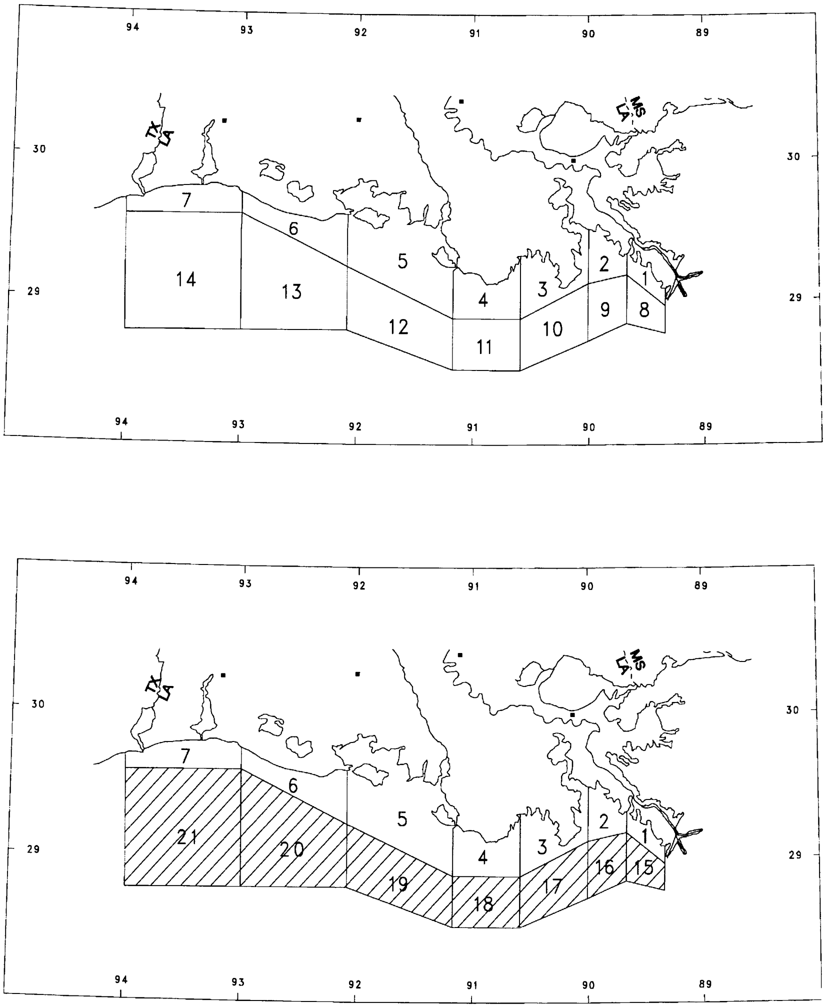

Figure 13. Model spatial segmentation grid for the NECOP water quality model (from Bierman et al., 1994; used with permission of the Estuarine Research Federation).

conditions arises because the model does not explicitly represent dissolved oxygen or nutrient processes in the sediments.

The principal water quality response parameters investigated were bottom-water dissolved oxygen concentrations and surface-water chlorophyll concentrations. All results for oxygen represent the average of Segments 15–21 for bottom offshore waters, and all results for chlorophyll concentration represent the average of responses for Segments 8–14 for surface offshore waters (Fig. 13). The results are expressed in terms of change relative to base calibration results.

Both dissolved oxygen concentrations (Fig. 14) and chlorophyll concentrations (Fig. 15) are responsive to nitrogen load reductions from the Mississippi and Atchafalaya Rivers. There is more variability in responses among different years for dissolved oxygen than for chlorophyll. As nitrogen loads are reduced from 10 to 70%, forecasted chlorophyll concentrations for the three years decreased by amounts ranging from approximately 1 to 72%. For the same nitrogen load reductions, forecasted dissolved oxygen concentrations for 1985 and 1988 conditions increased by amounts ranging from approximately 12 to 59%. In contrast, similar reductions for 1990 conditions increased the dissolved oxygen by approximately 28–148%.

Differences in responses of dissolved oxygen concentrations for 1990 conditions, as compared to 1985 and 1988, are probably due to differences in water

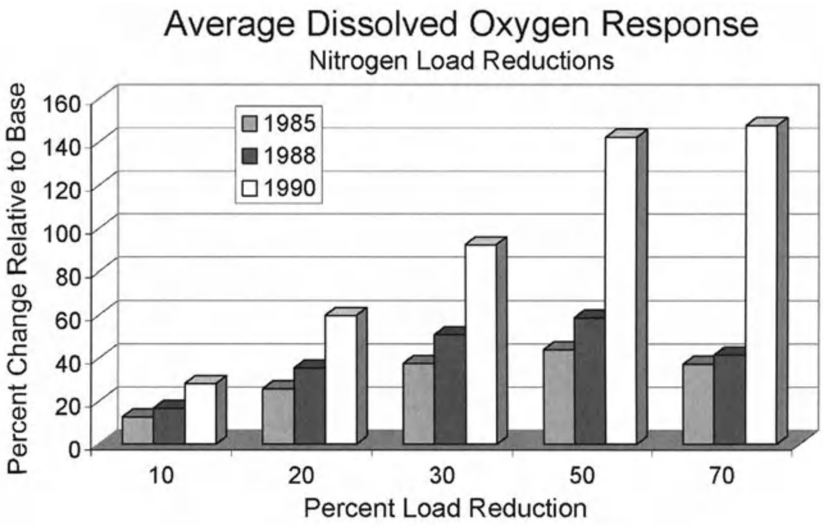

Figure 14. Forecasted responses of average dissolved oxygen concentrations (bottom offshore segments combined) to nitrogen load reductions from the Mississippi and Atchafalaya River system under 1985, 1988 and 1990 summer average conditions (from Bierman et al., 1999).

circulation patterns on the Louisiana inner shelf. Summer average conditions on the Louisiana inner shelf are typically represented by the Louisiana coastal current, which has a net westward drift along the shelf bathymetry. Typical summer average current speeds are approximately 10 and 3 cm s^{-1}, respectively, in the surface and bottom waters. During the summer of 1990, net eastward drift was observed in both surface and bottom waters at speeds of approximately 2 and 0.8 cm s^{-1}. It is possible that much smaller current speeds in 1990 affected the relative importance of physical transport *versus* chemical–biological processes for dissolved oxygen. Dissolved oxygen may have been affected more than chlorophyll because dissolved oxygen responses are more influenced by chemical–biological processes at the sediment–water boundary than are chlorophyll concentrations. In particular, at lower current speeds a parcel of water would be exposed to sediment oxygen demand for longer periods of time and hence would experience greater oxygen depletion than at higher current speeds. This result is important because it indicates that estimates of water quality responses to changes in nutrient loads can be strongly influenced by hydrometeorological conditions and water circulation.

An important management issue is the relative influence of nitrogen *versus* phosphorus loads in controlling bottom-water dissolved oxygen on the Louisiana inner shelf. Model forecast results indicated a general tendency for responses to be somewhat greater for nitrogen load reductions than for phosphorus load reductions (Fig. 16). As noted earlier, nitrogen is considered to be relatively more important than phosphorus in limiting primary productivity on the Louisiana inner shelf, but both phosphorus and silica may also be limiting at times and in certain locations, or combinations of nutrients may be limiting. Although nitrogen appears to be relatively more controlling than phosphorus in the NECOP water quality model and in forecast simulations, the computed differences are not large. Further investigation appears warranted for both these nutrients as well as the inclusion of silica into the water quality model. Field and experimental studies of nutrient limitation on the Louisiana inner shelf are continuing.

All of these forecast simulations represent steady-state responses, and provide no information on the time scales for potential water quality responses to nutrient load reductions. Over long-term seasonal data for station C6* (Fig. 1), Justic et al. (1993, 1997) estimated time lags of approximately one month between river discharge and nitrogen load and surface net production, and a subsequent lag of one more month for maximal bottom-water oxygen deficiency. Justic et al. (1997) further demonstrated that for years with large floods the organic carbon substrate laid down during the flood season may fuel the onset of hypoxia for more than a single year.

The ultimate response of bottom-water dissolved oxygen concentrations depends on the relative im-

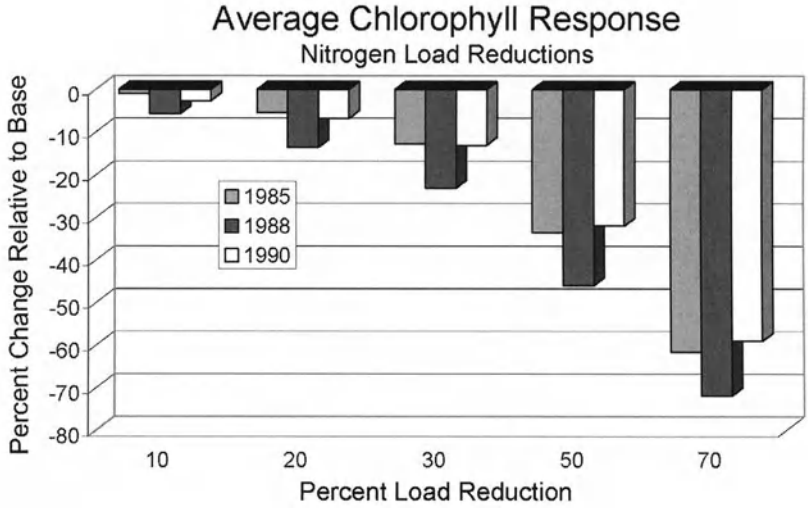

Figure 15. Forecasted responses of average chlorophyll concentrations (offshore surface segments combined) to nitrogen load reductions from the Mississippi and Atchafalaya River system under 1985, 1988 and 1990 summer average conditions (from Bierman et al., 1999).

portance of SOD versus water column processes in controlling bottom-water oxygen depletion rates. Total SOD consists of aerobic processes in the surficial sediments and anaerobic processes in deeper sediment layers. While surface layer processes may respond to loading changes on seasonal to annual time scales, processes in deeper sediment layers may take many years to respond. For example, forecast results from a coupled water-sediment mass balance model for Lake Erie (DiToro et al., 1987) indicated that the SOD component of total oxygen depletion rates did not reach steady-state until 5–10 years after changes in external nutrient loads. Results from a similar coupled model for Chesapeake Bay (Cerco, 1995) showed that decade-long simulations were required to achieve a near-complete response to external loading reductions.

Potential for change in the Mississippi River basin and the offshore marine ecosystem

The various modeling efforts for the Louisiana shelf indicate that hypoxia can be alleviated to some degree by a reduction in the nutrient loading. As demonstrated by models from other systems and the coupled biological–physical model of Justic et al. (1997), the response time of the marine system is unknown but may be several years or longer. The increase in nitrate loads within the Mississippi River watershed occurred over several decades, and it is possible that a new 'steady state' condition exists in the watershed based on stored nitrogen that responds to precipitation and climatic variability. Thus, the effects of any management actions undertaken to reduce nitrogen inputs to the basin may not be reflected in reduced nitrogen flux to the Gulf for a number of years. Subsequently, changes in the marine system in response to altered nutrient chemistry in the northern Gulf of Mexico may also lag behind nutrient load reductions.

An interagency Task Force of eight U.S. federal agencies and nine states and two tribal nations drawn from within the jurisdiction of the Mississippi watershed has proposed a plan to reduce the size of the hypoxic zone in the Gulf of Mexico and restore and protect the waters of the 31 states within the Mississippi basin. The plan proposes that the 5-year running average size of the Gulf of Mexico hypoxic zone be reduced to 5000 km^2 or less by the year 2015 through a series of voluntary and incentive-based activities, mostly within the watershed, that would reduce the nitrogen load to the Gulf of Mexico by 30% of the average 1980–1996 level, on a similar running average and time frame. These efforts will take place within a larger framework of increasing demand for agricultural products from the continental United States and a variable climate. Climate, in the form of variable precipitation, is a significant influence on both the nutrient loads and the offshore stratification. River discharge varies, but the long-term Mississippi River discharge values back to the 1800s show no long-term increases in discharge. Nitrate concentrations, on the other hand, have increased over that period, particu-

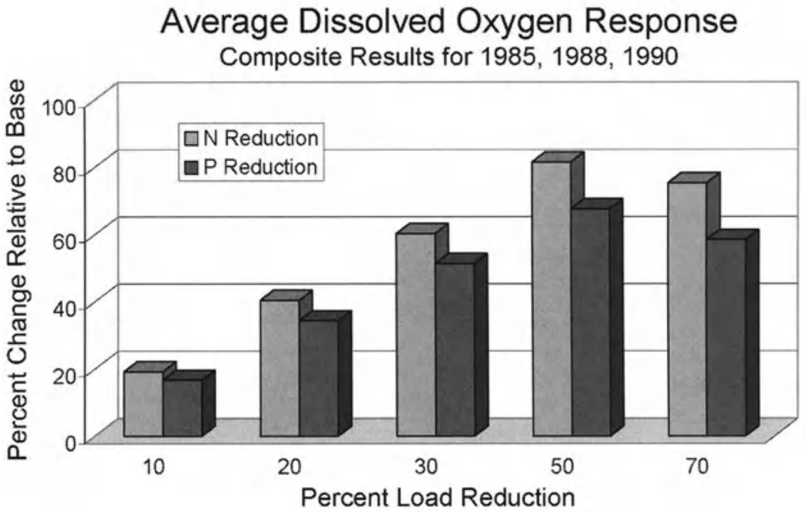

Figure 16. Forecasted responses of average dissolved oxygen concentrations (bottom offshore segments) to nitrogen and phosphorus load reductions from the Mississippi and Atchafalaya River system composited for 1985, 1988 and 1990 summer average conditions (from Bierman et al., 1999).

larly in the spring period when discharge is higher, so that the total nitrogen load has subsequently increased. Nutrient management activities and landscape alterations that reduce the flux of primarily dissolved nitrogen from the watershed are necessary for a reduction in the total nitrogen load to the Gulf of Mexico.

Similar and more restrictive goals for overall nutrient reduction to coastal systems have been proposed, initiated and even legislated for other areas of the United States and the world (Boesch & Brinsfield, 2000; Boesch et al., 2001). Nitrogen reduction goals of 40% and 58.5% have been set for the Chesapeake Bay and Long Island Sound, respectively, and represent some portion of the total load deemed controllable, rather than a percentage of the total load. Nitrogen load reductions of 50% have been set for both the Baltic and North Seas. In each of these areas, through numerous activities, load reductions have been achieved. The manifestation of these reductions have been observed in improvements in water quality, and, in some cases, improvement in environmental indicators. On smaller scales than the Mississippi River watershed and Gulf of Mexico hypoxic zone, the detrimental effects of eutrophication have been mitigated by reduced nutrient loads (Rabalais, 2002). A major watershed, still smaller than that of the Mississippi basin, contributes to the nutrient load of the Black Sea where an area of hypoxia as large or larger than that of the northern Gulf of Mexico has existed since the early 1970s as a result of eutrophication (catchments of 1.4 $\times$ 10^6 km^2 and 2.9 $\times$ 10^6 km^2 for the Danube and the Mississippi, respectively, and hypoxic zones of up to 20 000 km^2 for each area) (Tolmazin, 1985; Zaitsev, 1992; Mee, 2001). In the Black Sea catchment, the use of phosphorus and nitrogen fertilizers in the Danube and Dnieper Rivers fell dramatically with the collapse of a centralized Soviet economy in 1989–1991 and resulted in substantial reductions in the discharge of nitrogen and phosphorus into the Black Sea. The zone of severe hypoxia on the northwestern shelf of the Black Sea disappeared in 1996 for the first time in 23 years and has remained small ($<$1000 km^2 in 1999 for the Black Sea shelf). The lesson to be learned from other areas where the time sequence towards eutrophication escalated since the 1950s with concomitant degraded coastal ecosystems and where nutrient reduction and retention strategies have reduced loads to coastal regions, is that nutrient management can result in improved water quality of marine habitats. With the commitment of individual, societal and political will, reverses in the increasing flux of nitrogen from watersheds and worsening coastal water quality can be accomplished.

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Potential modification of the fluxes of nitrogen from the Humber Estuary catchment (U.K.) to the North Sea in response to changing agricultural inputs and climate patterns

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Key words: Humber, Ouse, Trent, catchment, river, estuary, models, nitrogen

Abstract

The catchment of the Humber Estuary drains approximately 20% of the land area of England via two main rivers, the Trent and the Ouse, and a number of tributaries. The catchment is home to major metropolitan and industrial centres, as well as to extensive areas of agricultural land; for this reason, the river and estuarine systems have been subject to considerable anthropogenic inputs. The Humber Estuary is one of the largest U.K. estuaries and the major U.K. freshwater input to the North Sea. The U.K. Natural Environment Research Council (NERC) Land Ocean Interaction Study (LOIS), which combined extensive physical and biogeochemical measurements with an integrated modelling programme, was established to examine the transport and fate of nutrients and other constituents through the land-sea boundary. In this paper, a model of nitrogen (nitrate, nitrite, ammonium, particulate nitrogen) transport and cycling in the Humber Estuary, calibrated on the basis of measured constituent concentrations at its riverine and marine boundaries, is linked off-line to a Humber catchment and rivers model of nitrogen transport, which furnished simulated constituent values at the tidal limits, and the resulting estuarine nitrogen profiles compared to those of the standalone estuarine model. The estuarine model is then re-run using simulated concentration values at the tidal limits from catchment-river model simulations incorporating realistic changes in agricultural fertiliser inputs and climate forcing functions. The standalone estuarine model simulation estimated nitrate+nitrite ($\sim$total nitrogen) export to the North Sea to be ca. 53 000 t in 1994 and 44 000 t in 1995. Following linkage of the estuarine and catchment-river models, the estimated fluxes for these years increased by 20–30%, relative to the standalone simulation. Higher winter riverine flows largely accounted for this difference. The altered flows also markedly changed the simulated concentrations and distributions of suspended particulate matter (SPM) within the estuary, indicating strongly that the transport and fluxes of particle-reactive and particle-associated constituents would show measurable differences. Scatter in the measured SPM data precluded identification of the more precise simulation run, however. Subsequent simulations using the linked models estimated that a 50% reduction in artificial fertiliser applications within the catchment gave a 10–15% decrease in nitrogen loads to the North Sea, relative to the 1994–95 input, whilst forcing the catchment model with a climate perhaps appropriate for the mid-21st century yielded nitrogen fluxes that were similar to those of the mid-1990s.

Introduction

The U.K. NERC LOIS programme was an interdisciplinary study designed, inter alia, to improve the understanding of the land to sea fluxes of materials and to do so in a manner that will be of use to environmental managers and policy makers. An important product of LOIS has been the generation of an extensive terrestrial, estuarine and marine database which has contributed greatly to the development of a series of biogeochemical transport models for use in these environments. A key goal in the latter stages of LOIS has been to link this suite of models so that the present day fluxes of contaminants and other materials from the river catchment to the open ocean can be estimated. Subsequently, the intention has been to use the models to investigate the potential effects, on the estimated fluxes, of selected realistic scenarios of climate change and land use over the next 50–100 years (Stebbing et al., 1998).

This paper describes the current status of three models that, between them, describe material transport in the catchment, rivers and estuaries of the Humber watershed, with particular emphasis on the estuarine component of the system. Representative simulations of the estuarine profiles of salt, SPM and of nitrogen species (principally nitrate and nitrite) based on measured freshwater flows and concentrations, which were generated during the calibration phase of the estuarine model, are reported, together with measured estuarine values for these variables and estimated fluxes to the North Sea. The river-catchment and estuarine models are subsequently linked off-line and the contemporary simulation is repeated, and the resulting estuarine profiles and fluxes to the North Sea compared. These simulations are referred to as the standard and baseline runs, respectively. The linked models are then used to examine how changes in land use and climate within the catchment may alter the estimated nitrogen fluxes to the marine environment.

Site description

The catchment area draining to the Humber Estuary is approximately 24 000 km^2 (~20% of the land area of England) and it delivers a mean freshwater flow to the North Sea of ca. 250 m^{3}s^{-1}, the largest from any estuary on the U.K. east coast (Jarvie et al., 1997). Several large, industrially important, metropolitan areas are located within the catchment, together with extensive areas of farmland; as a result the rivers of the catchment have historically received substantial anthropogenic inputs. Probably for this reason, nutrient concentrations in some of the rivers are high, often exceeding 10 mg l^{-1} nitrate and 3 mg l^{-1} ammonium, depending on flow conditions (Jarvie et al., 1998). The Humber Estuary is formed by the confluence of two rivers, the Trent, which drains the southern part of the Humber basin, and the Ouse, which drains the northern and western parts (see Fig. 1). The River Ouse catchment is divided into five sub-basins, the Don, Aire, Wharfe, Ouse and Derwent. Together, the six catchments represent 71% of the drainage area and 90% of the freshwater flow to the estuary. An overview of the geography of the region, and of the water quality of the rivers, is given in Jarvie et al. (op. cit.) and House et al. (1997).

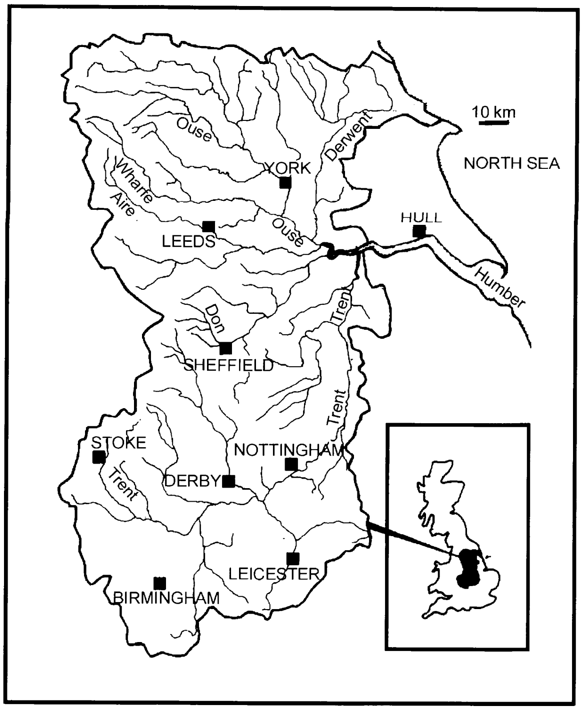


Figure 1. The main river systems of the catchment of the Humber Estuary. Modified from Rees et al. (1998).

The estuarine part of the system extends from Spurn Head on the North Sea coast to Naburn Weir on the Ouse and to Cromwell Lock on the Trent. The tidal reaches of the Ouse and Trent, which are 61 km and 85 km long, respectively, converge at Trent Falls, where they flow into the Humber. The Humber itself is 63 km in length, and is macrotidal, with spring and neap tides at the mouth of 5.7 m and 2.8 m, respectively. As a consequence of tidal stirring, the estuary is vertically well-mixed with respect to salinity. It is also highly turbid, particularly in the vicinity of Trent Falls where surface suspended matter concentrations can reach 20 g l^{-1} during the summer months (Uncles et al., 1998a). Like the catchment rivers, the Humber Estuary and tidal Ouse have also received substantial inputs of sewage and industrial effluent (National Rivers Authority, 1993).

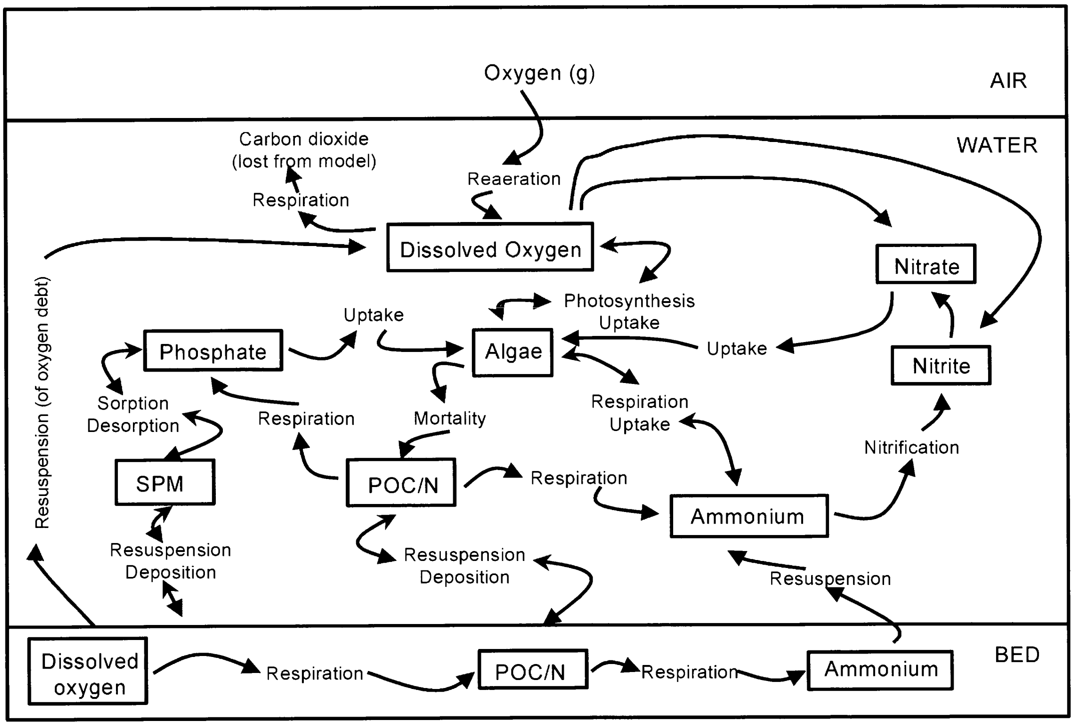


Figure 2. State variables, and their transformations, included in the estuarine water quality model. POC is particulate organic carbon, SPM is suspended particulate matter.

Model descriptions

The catchment and river models are dynamic, and incorporate process-based descriptions of the transport and transformations of water, SPM, nitrogen and other constituents. Both models have been extensively described elsewhere (Cooper & Naden, 1998; Eatherall et al., 1998; Proctor et al., 2000; Tappin et al., submitted), and only an outline of their characteristics is given here.

The catchment delivery model represents the land surface in terms of hydrological response units (Cooper & Naden, op. cit.) and incorporates a spatially distributed representation of processes. Data requirements of the model include information on climate (rainfall, temperature and potential evapotranspiration), land form (elevation and soil types) and land-use (including agricultural management and the use of fertilisers).

The river model ('QUESTOR', Quality Evaluation and Simulation Tool for River Systems; Eatherall et al., op. cit.) is a 1-D formulation that represents the major channel network of the catchment, and simulates in-channel advection and dispersion of constituents and also their transformations. The state variables within QUESTOR include nitrate, nitrite, ammonia/ammonium and particulate nitrogen. The inputs to this model are the outputs from the catchment model and direct discharges to the streams and rivers from point sources (sewage effluent, industrial discharges). The river model has been calibrated for the period 1986–90, and tested over the years 1991–95, using data from the Environment Agency of England and Wales, both at sites near to the tidal limits and at those located within upstream catchments. Both the catchment and river models use a daily time step, which is adequate for representing the dynamics of the systems and the variability in fluxes over extended periods. The tidal limit of each the Humber rivers is defined by either a weir or a lock, thus providing a clear boundary between the river and estuarine models. There is no advection or diffusion of material from the estuary in to the rivers across these boundaries.

The simulated tidal system is represented by the Humber, extending from the confluence of the Ouse and Trent estuaries at Trent Falls to Spurn Head on the North Sea coast; the Ouse Estuary, extending from the tidal limit at Naburn Weir, and the Trent Estuary, extending from the tidal limit at Cromwell Lock. Fluxes (in kg d^{-1}) of constituents into the estuary across the tidal limits are calculated from river flow (in m^3 d^{-1}) and constituent concentration (in kg m^{-3}). Each simulated branch is 1-D, with variability restricted to along its axis. The model is tidally averaged to enable the study of the seasonal and annual variations in the transport and transformations of constituents. The advection and dispersion of salt, SPM, nitrate, nitrite and ammonium are simulated, as well as other variables. Particulate nitrogen is not explicitedly represented, although it can be derived from particulate organic carbon which is dynamically simulated. Water velocity is determined by cubature, and the mixing of solutes is a function of water velocity and axial dispersion. SPM is represented by both permanently and temporally suspended particles, the latter exchanging with the estuarine bed through cycles of deposition and resuspension. The state variables, and their transformations, incorporated in the model are shown schematically in Figure 2.

The model has been calibrated using near-surface data from the 27 axial surveys of the Humber and tidal Ouse collected between 1994 and 1996 as part of the LOIS programme (Uncles et al., 1998a, b, c). The tidal Trent was not as extensively sampled, and so simulations of this system incorporate parameters and their values that have been estimated for the Ouse and Humber. The estuarine model has been constructed using the ECoS (version 3) simulation software (Harris & Gorley, 1998a,b; Gorley & Harris, 1998). A more extensive description of the model and its calibration and testing is given elsewhere (Tappin et al., submitted), and an overview is given in Tappin et al. (2001). For additional examples of ECoS based estuarine models, the reader is referred to Pham et al. (1997) and Liu et al. (1998).

Scenario descriptions

The U.K., in concert with other European countries, is obliged by international agreement to reduce its inputs of nitrogen to the marine environment in order to counter problems of eutrophication. For example, the OSPAR Convention of 1992 set the goal of reducing nitrogen inputs to coastal waters by 50% over the period 1985–95 (Oslo & Paris Commissions, 1992). More recently, the European Union Directive on the Protection of Waters against Pollution caused by Nitrates from Agricultural Sources (the 'Nitrates' Directive) has been specifically enacted in order to reduce the impacts of agricultural runoff on coastal eutrophication. In response to requests from the U.K. Environment Agency of England and Wales and the Ministry of Agriculture, Fisheries and Food, the models have been used to examine the contribution of farm fertiliser applications to nitrogen fluxes from the Humber Estuary to the North Sea. For this scenario, the catchment model was run with artificial fertiliser applications reduced by 50% relative to the late-1980s and early-1990s.

Also in response to requests from U.K. statutory agencies, the models have been used to investigate how nitrogen fluxes to the North Sea may change as a result of altered meteorological conditions. The new climate regime was taken from the results of simulations carried out by the U.K. Climate Impacts Programme (Hulme & Jenkins, 1998). The modelled scenario, known as the HADCM2 Medium High Scenario, is based on the change in climate being forced by a 1% per annum increase in greenhouse gases over the next century. Mean conditions simulated for the years 2040–2069 have used to drive the catchment model. Relative to the average for the period 1961–90, the climate simulations suggest that the mean temperature will be 2 °C higher, mean rainfall 15% higher (and with a marked increase in the summer-winter contrast in rainfall) and potential evapotranspiration 3% greater (again with a markedly increased seasonal contrast).

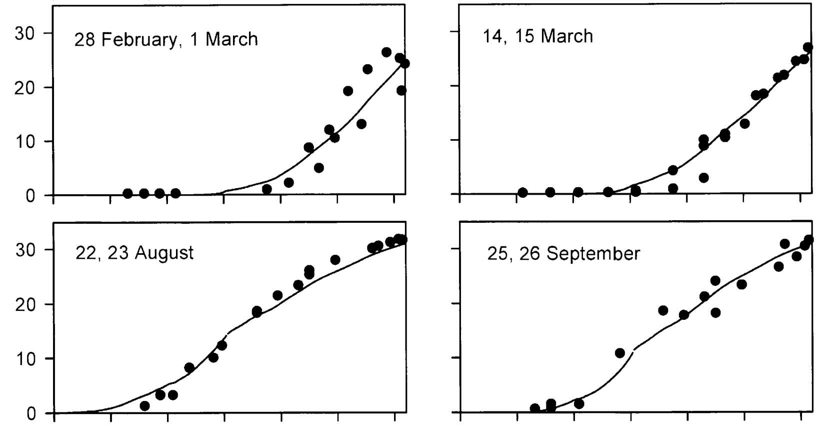
For the scenario simulations, the catchment-river models are run for the years inclusive 1986–1995 and the estuarine model from mid-1993 to the end of 1995, thus giving a run-up period of 7.5 years for the estimation of the boundary conditions at the tidal limits.

Results and discussion

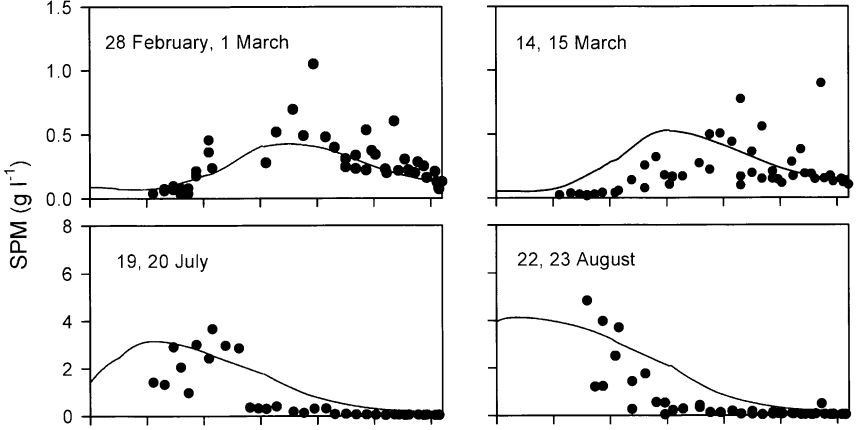
Estimates of the present day fluxes of nitrogen

It is not the intention to extensively reproduce the results from the calibration of the estuarine model here, as these are to be reported elsewhere, as noted above. However, there are marked differences between the estuarine water flows, axial profiles of modelled constituents, and thus their exports to the North Sea, when generated from the standard and baseline runs for certain constituents, implying that currently there are constraints to the use of linked models for estimating land to sea fluxes, at least in this system. Representative results from the standard simulations are reported to illustrate this finding. Axial profiles

(a) Salinity



(b) Suspended particulate matter



(c) Nitrate+nitrite

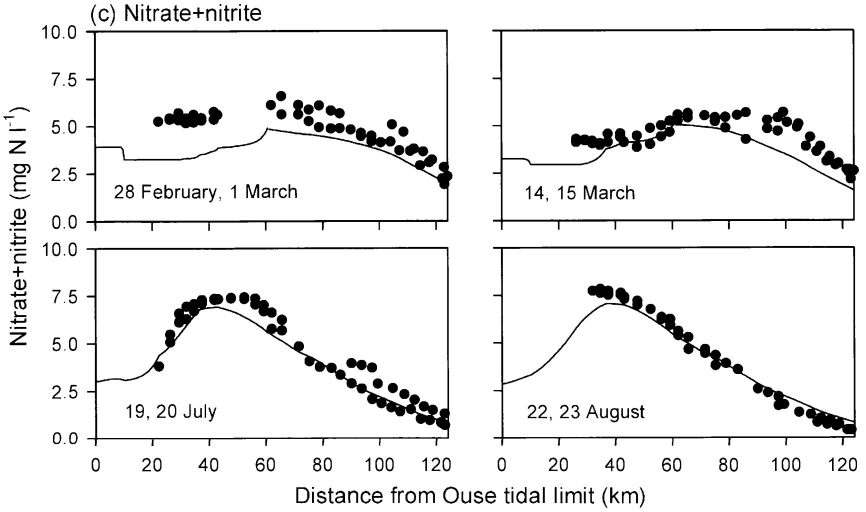


Figure 3. (a) Observed (circles) and simulated (solid line) salinity within the Ouse-Humber estuaries for 1995. (b) Observed (circles) and simulated (solid line) suspended particulate matter within the Ouse-Humber estuaries for 1995. (c) Observed (circles) and simulated (solid line) nitrate+nitrite within the Ouse-Humber estuaries for 1995. The observations are near surface values, whilst the simulated data are from the sectionally and tidally-averaged estuarine model.

from 1995 have been selected for inclusion from the larger number available, although flux information for both 1994 and 1995 are reported.

The simulated salinity profiles show a good fit to those observed between 1994 and 1996, during which wide ranges in flow and tidal conditions were encountered. Typical examples, from February to March and August to September, representing high and low river flow conditions, respectively, are shown in Figure 3(a). Inevitably, for SPM there was a large scatter in the observed concentrations due to differences in the accretion, erosion and mixing of different particle types (unlike salinity and other solutes), coupled to marginally asynoptic sample collection (although typical of high water when concentrations are greatest) (Uncles et al., 1998a). Nevertheless, the model does clearly reproduce the order of magnitude variations in turbidity between surveys, as well as the general position and size of the turbidity maximum, as exemplified in Figure 3(b). However, the simulation does generate higher turbidities in the upper reaches of the Ouse than were observed, and in a number of cases places the turbidity maximum rather more up-estuary than the observations suggest. The latter phenomenon is not reproduced in the Trent Estuary, and this aspect of the model simulations requires further examination.

Examples of observed and simulated axial profiles of nitrate+nitrite are shown in Figure 3(c), although nitrite contributes only a few percent to the total. For nitrate, there are effluent inputs and non-conservative addition and removal (nitrite oxidation, plankton uptake, denitrification), but quantitatively these factors and processes are not significant in the Humber Estuary, and to a first approximation, nitrate can be considered as a conservative tracer between river waters of high nitrate concentrations and seawater of lower concentration. This behaviour largely accounts for the good fit for nitrate+nitrite between the simulated and observed values. On a number of occasions, however, there is a systematic under- or overestimation of the simulated concentrations with respect to the observed values, particularly in the region spanning the lower Ouse and upper Humber estuaries. The inflows of the Aire, Don and Trent are located within this region and the discrepancies between the observed and simulated data can be largely explained by inaccuracies in the estimation of the river loads. This is because a log-transformed least squares regression between river flow and constituent concentration was used to calculate daily concentration data from observational data of lower resolution (weekly or less). The introduction of errors in this way is unavoidable when using infrequent sample data, although the magnitude of the error can be mitigated by the use of the most appropriate algorithm from one of the many interpolation and extrapolation routines available (Phillips et al., 1999; Webb et al., 2000). The discrepancy between the modelled and observed SPM in this region does not contribute to any extent to the differences in simulated and measured nitrate+nitrite, via ammonification of particulate nitrogen and subsequent nitrification of the ammonium, because the concentrations of reduced nitrogen species are small compared to those of nitrate.

Table 1. Estimated fluxes of nitrate and nitrite from the Humber Estuary to the North Sea for 1994 and 1995[a] (t a^{-1})

Model simulation	1994		1995	
	Nitrate	Nitrite	Nitrate	Nitrite
Standard	52 700	480	43 200	520
Baseline	68 400	280	51 400	180
Fertiliser reduction	61 500	280	43 400	180
Climate change	67 300	370	47 800	230

[a] The results from the climate change simulations are hypothetically representative of the period 2040–2069.

Table 1 provides a summary of the export of nitrate and nitrite during 1994 and 1995. The values for their export estimated from the standard run are in good agreement with the value of 55 200 t a^{-1} calculated by Sanders et al. (1997) for combined nitrate, nitrite and ammonium in the early 1990s. Ammonium and particulate nitrogen have not been included in the Table because their export is trivial relative to nitrate+nitrite, and during 1995 the estuary appeared to be a minor sink for both constituents. For ammonium, this can be explained by extensive nitrification within the estuary (Barnes & Owens, 1998).

Comparisons of the model output from the standard (measured river inputs) and baseline (simulated river inputs) simulations show that differences between the axial distributions are relatively small for salinity and nitrate+nitrite but are more marked for SPM (see Fig. 4). Thus, during the winter, high flow periods, concentrations of SPM from the standard run are systematically higher than those from the baseline simulation. In summer the converse is the case, with markedly higher concentrations generated from the baseline simulation compared to the standard output, particularly within the turbidity maximum zone. These trends are also evident in the Trent Estuary simulations. The differences in the SPM profiles arise

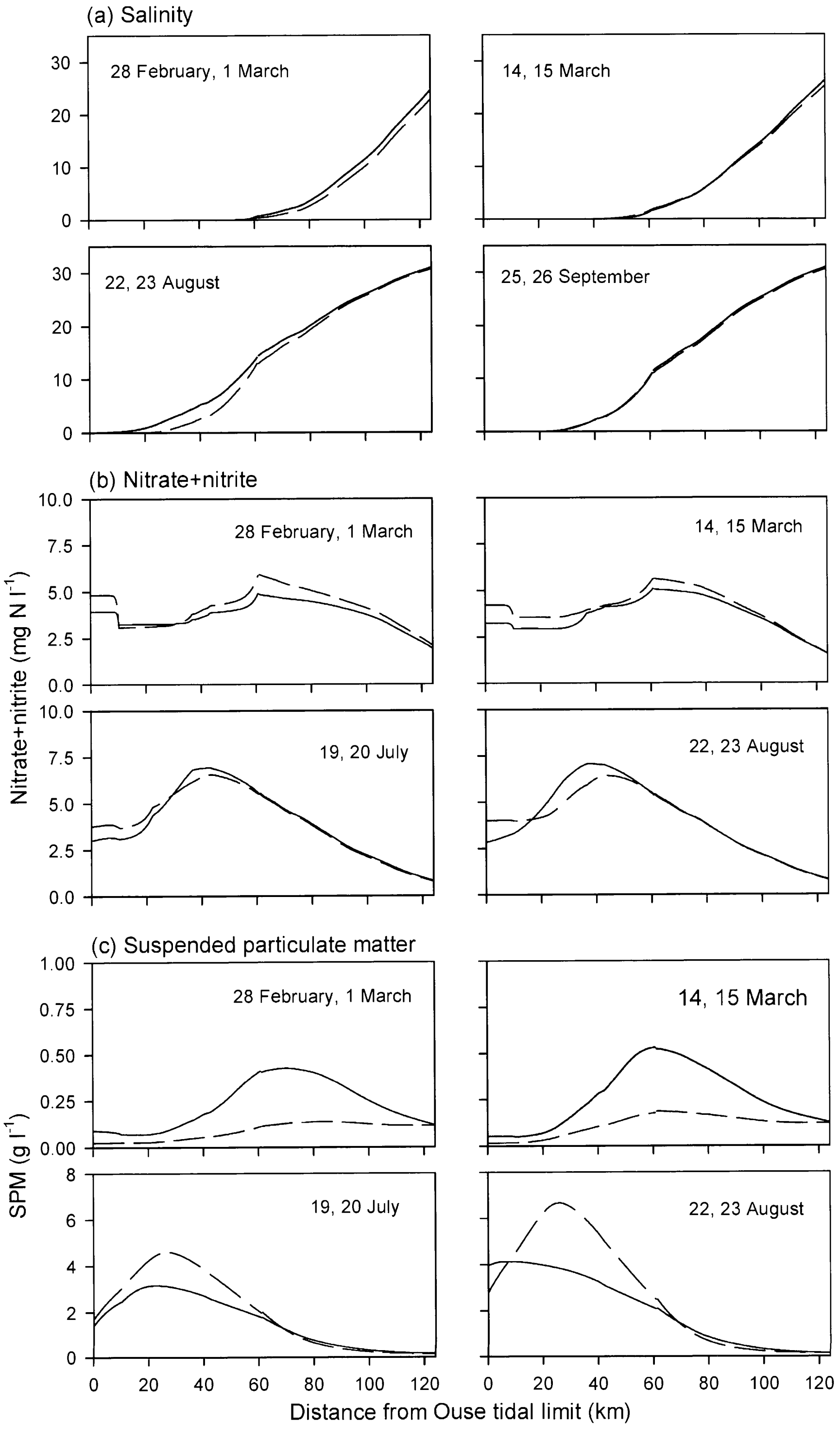


Figure 4. (a) Simulated salinity within the Ouse-Humber estuaries for 1995. (b) Simulated nitrate+nitrite within the Ouse-Humber estuaries for 1995. (c) Simulated suspended particulate matter within the Ouse-Humber estuaries for 1995. The results were obtained from the sectionally and tidally-averaged estuarine model. Standard run (solid line), baseline run (dashed line).

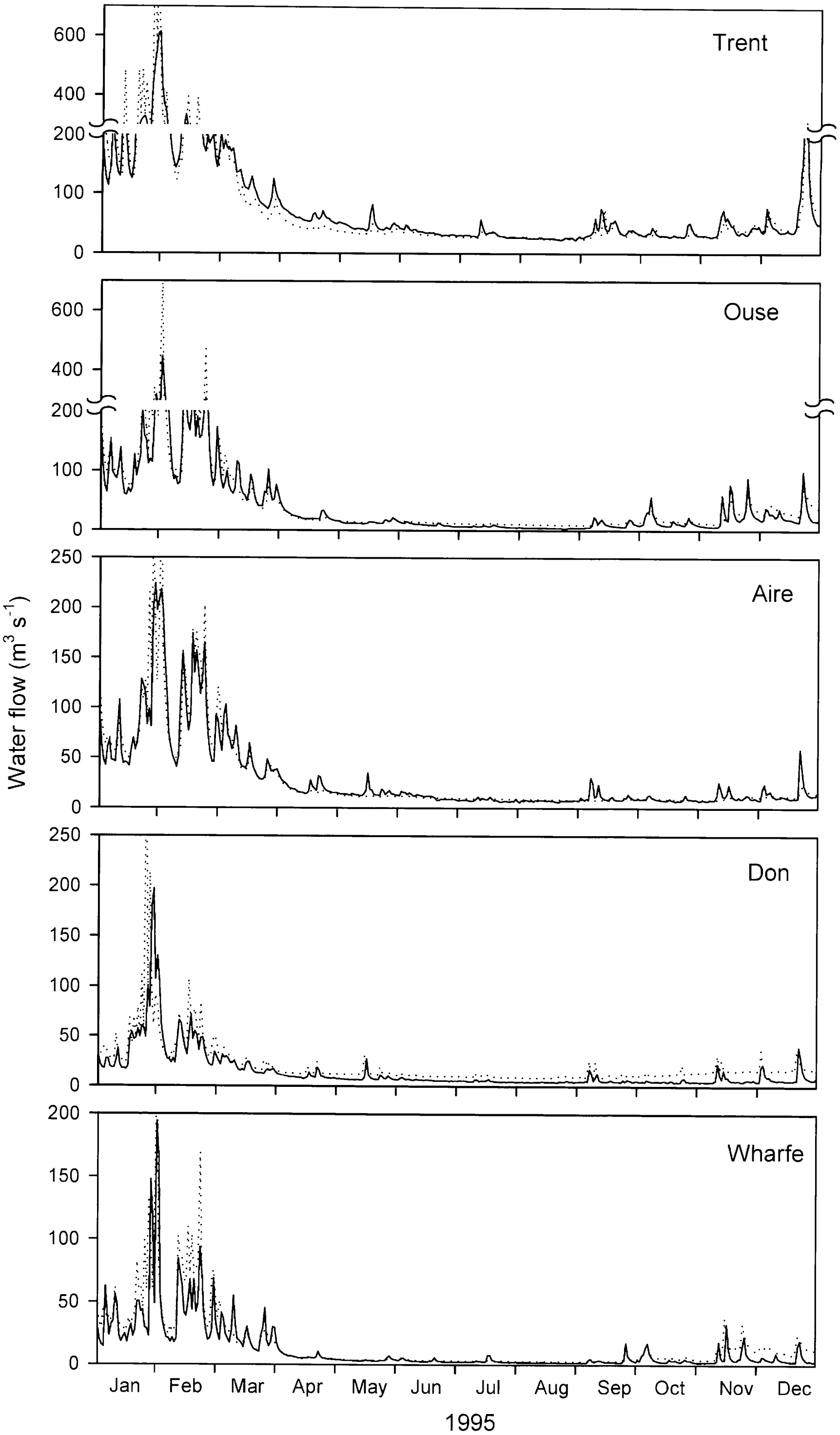


Figure 5. Flows of river water, at the tidal limit, into the estuarine model from the major Humber catchment rivers and tributaries during 1995. The measured values (solid line) totalled 165 292 m^3 and the baseline simulated values (dotted line) 192 099 m^3 over the 2 year period.

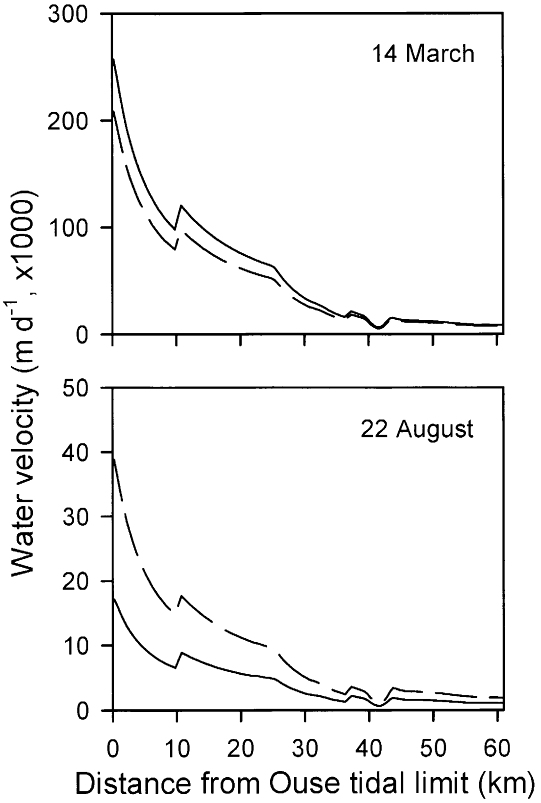


Figure 6. Simulated water velocities in the Ouse Estuary during two representative periods from 1995. Standard run (solid line), baseline run (dashed line).

because the flows of particles within the estuary are convergent, and concentrations are therefore sensitive to both riverine and marine boundary conditions.

Numerical experiments have shown that the apparently minor differences in the measured and modelled river water inflows at the tidal limits, particularly during the spring and summer months, as shown in Figure 5, give rise to differences in water velocities within the estuary, as indicated in Figure 6. These, thereby, change the particle transport in the estuary, and cause the marked differences in the SPM distributions. The higher concentrations of suspended particles generated during the summer by the baseline simulation are consistent with the greater mass of SPM in the estuary calculated for April–September 1995, and also consistent with the reduced fluxes of SPM to the North Sea during the spring of 1995, relative to the standard run, as exemplified in Figure 7. Model experiments also showed that the minor differences in SPM concentrations at the tidal limits had some effect on estuarine concentrations and distributions of SPM, although the effect was negligible during the summer when turbidities in the estuary were high.

Whilst the differences in water flows within the estuary have little effect on the axial profiles of salinity and nitrate+nitrite, they do effect the actual fluxes of water and nutrients to the North Sea. This can be seen in Table 1 where the nitrogen export is 20–30% higher from the baseline simulation.

Because of the scatter in the observed data for SPM, it is likely that the baseline simulation lies well within the margin of error associated with the standard simulation. However, the inference is that, relative to the estimates for conservative solutes, there will be even less agreement between the flux estimates from the standard and baseline combinations for solutes whose concentrations are measurably influenced by particles (e.g. ammonium, dissolved oxygen) or for constituents that are primarily associated with particle matrices (e.g. hydrophobic micro-organic contaminants). This is not only because the particle concentrations are dissimilar, but also because residence times, and hence the extent of particle-water interactions, will differ.

Scenario simulations and fluxes

Despite the caveats outlined in the preceding section regarding the precision of the flux estimates, model simulations incorporating the effects of fertiliser reductions and climate change have been made in order to understand how fluxes of nitrogen may change in a relative way. The results show that for all constituents, differences in the axial concentration profiles from the baseline and scenario runs are relatively minor, although larger for the SPM profiles resulting from the climate change scenarios. However, the simulated concentrations remain within the scatter of the observed data, and thus within the uncertainties of the model as currently formulated. Examples of the baseline and scenario generated estuarine profiles of SPM and nitrate+nitrite for contrasting winter and summer periods are given in Figure 8. The estimated exports of nitrate and nitrite to the North Sea resulting from effects of climate change, given in Table 1, show only minor differences relative to the baseline estimates. There is a reduction in nitrogen export following reductions in artificial fertiliser inputs, although the decrease is only 10–15%.

The apparently limited reduction in the export of nitrate following the reductions in fertiliser applications is perhaps surprising given the importance of

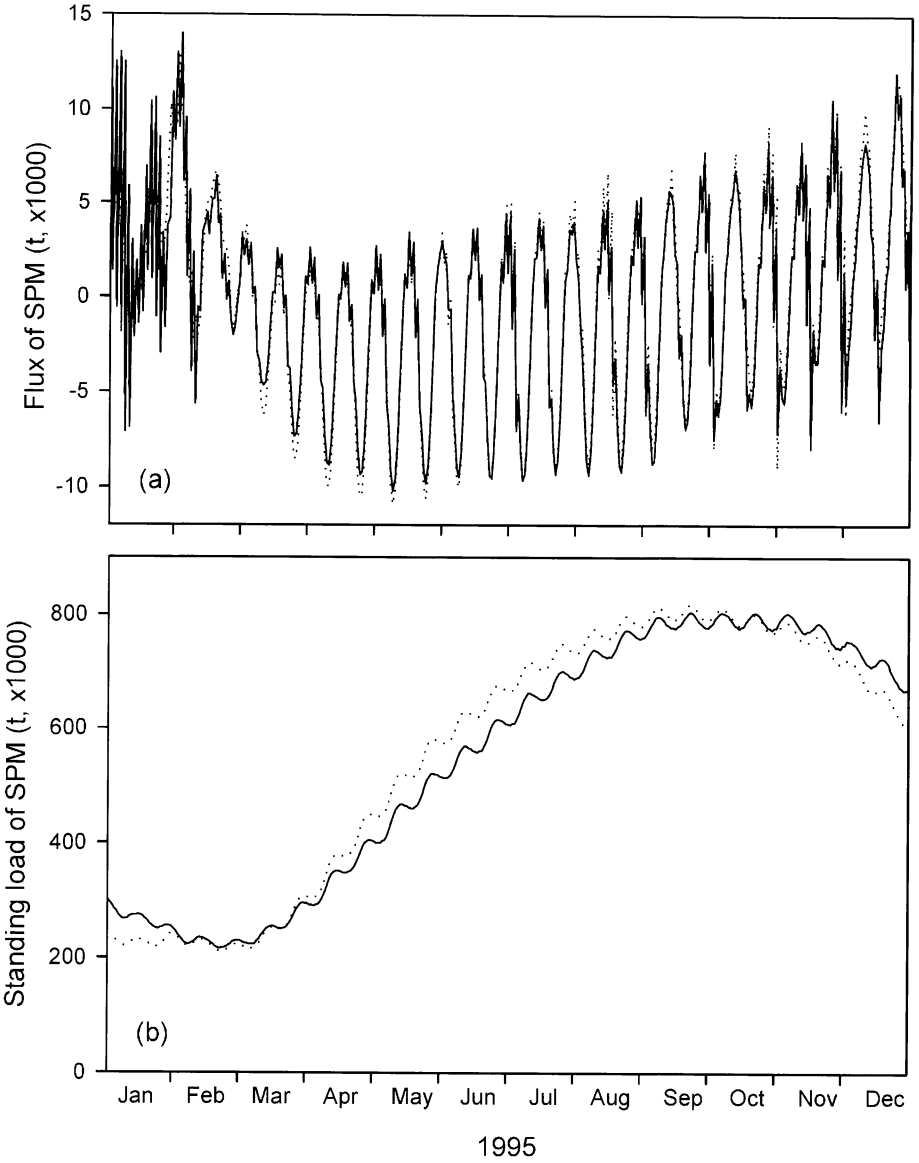
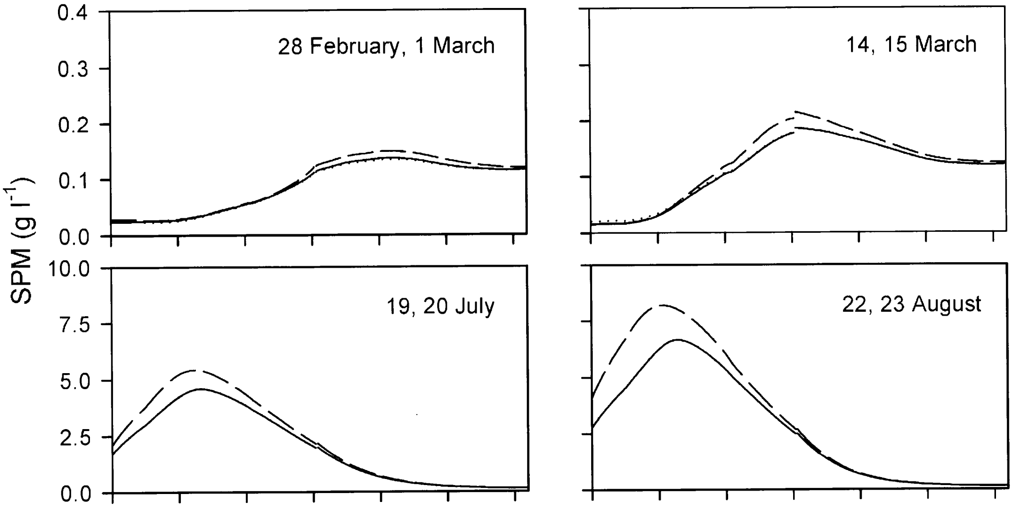


Figure 7. (a) Simulated fluxes of suspended particulate matter at the North Sea boundary (Spurn Head) during 1995. Export to the North Sea (+ve values), import from the North Sea (-ve values). (b) Simulated standing load of suspended particulate matter within the Humber, Ouse and Trent estuaries during 1995. Standard run (solid line), baseline run (dotted line).

agricultural runoff in raising river nitrate concentrations over the last 40 years (European Environment Agency, 1995). However, within the Humber catchment the amount of nitrogen applied in artificial fertilisers is less than half the total nitrogen fertiliser applied (i.e. artificial plus animal manure) to agricultural land (European Environment Agency, 1995). It is also the case that the nitrogen applied in artificial fertilisers is efficently removed by the crops on which it is used, implying that a reduction in applications simply leads to reduced uptake by the crop plants, rather than a 1:1 reduction in leaching to the rivers. The absence of a reduction in nitrogen loads is also consistent with the nitrate concentration–flow relationships reported for the Trent, Aire, Don and Wharfe, which suggest that the high nitrate concentrations in these rivers (and ammonium in the Aire and Don) largely reflect inputs from point sources, including sewage and industrial effluent. Only for the Ouse and Derwent are nitrate concentrations clearly influenced by agricultural run

(a) Suspended particulate matter



(b) Nitrate+nitrite

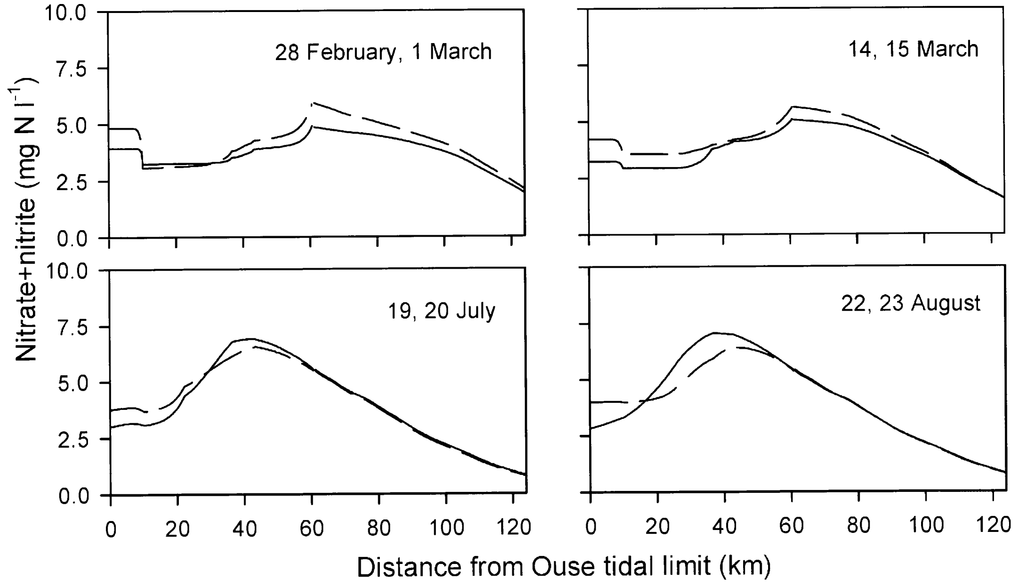


Figure 8. (a) Simulated suspended particulate matter within the Ouse-Humber estuaries for 1995. (b) Simulated nitrate+nitrite within the Ouse-Humber estuaries for 1995. Results obtained from the sectionally and tidally-averaged estuarine model. Baseline run (solid line), fertiliser reduction run (dotted line, obscured by solid line), climate change run (dashed line).

off (Jarvie et al., 1998). For this system then, it would appear that significant reductions in nitrogen loads to the North Sea will only be achieved by decreases in other nitrogen sources.

The fluxes resulting from the climate change scenario appeared to be similar to those estimated for the present day, as noted above. However, the simulation did not take into account, inter alia, any potential change in the morphology of the estuary through variations in mean sea level. It has been estimated that by 2050 there will have been a rise in sea level along the adjacent East Anglian coast of 37 cm, of which 28 cm will be due to climate change (I. Shennan, in Hulme & Jenkins, 1998). Through a hypothetical policy of managed realignment along the Humber coast following these increases in sea level, and the subsequent expansion of sub-tidal and inter-tidal mudflats, and saltmarsh, Jickells et al. (2000) have calculated that the retention of riverine nitrogen inputs through trapping or denitrification would increase by approxim-

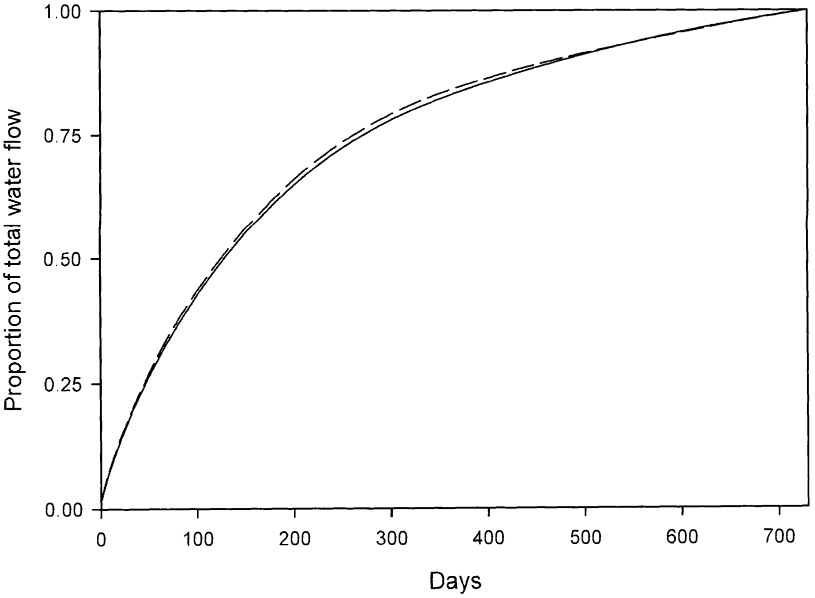


Figure 9. Temporal increase in the amount of river flow from the Humber catchment rivers into the Humber Estuary, as a proportion of the total flow, for the years 1994–95.

ately 15 times relative to contemporary fluxes (from 4% to 58%). Thus, the most significant climate induced alteration to nitrogen fluxes may arise through marked changes in estuarine morphology and function, rather than through changes in fluxes from the Humber catchment itself.

Summary and conclusion

This study indicates the potential of numerical models to quantitatively describe the natural environment, particularly when model construction and testing is allied to spatially and temporally extensive, high quality data. A calibrated model of the Humber estuarine system gave estimates of the flux of nitrate and nitrite to the North Sea during 1994 and 1995 of approximately 44 000 to 53 000 tonnes per annum, which accounted for practically all of the nitrogen exported.

At the same time, however, the study illustrates some of the problems inherent in the estimation of fluxes, particularly in complex non-linear systems of this sort. When, instead of being driven directly by observed river flows, the estuarine model was linked to a sophisticated and calibrated model of the river catchments and the rivers that feed the estuary, the estimated fluxes of nitrogen to the North Sea were increased by 20–30%. This was largely the effect of small proportionate errors in the estimated river flows during periods of high flow, as shown in Figure 5. These errors had negligible effect on the distribution of an inert solute such as salinity in the estuary, but, as the variation in river flow (and nitrate load also, in this case) tends to be proportional rather than additive, as indicated in Figure 9 where 50% of the total water flow occurred in only 19% of the time, they had a marked effect on the (additive) fluxes. In observational studies to determine fluxes of this kind, the problem that this illustrates is usually identified as the difficulty of monitoring the rare spate events that are recognised as carrying large proportions of the flux. The problem, however, is inherent in attempting to use essentially arithmetic measures (average fluxes) to characterise proportional variation in the instantaneous flow of material.

The use of completely modelled flows also altered the total mass of particles held in suspension by the simulated estuary, which led to the development of marked differences in the concentrations of suspended particulate matter, particularly during periods of low flow. This difference would perhaps be sufficient to markedly affect the distribution and flux of particle-reactive and particle-associated constituents, and the intensity of particle-associated activities such as nitrification and deoxygenation (Owens, 1986). Thus the particle dynamics appear to effectively amplify uncertainties in flow whose effect on the distribution of inert solutes is negligible. The mechanism(s) contributing to this effect is not introduced by the model but is inherent in the dynamics of the particles in the estuary, as indicated by the very large variation (both clearly through space and time, and allocated to 'error' in the model) in observed particle concentrations.

Both of these problems can be ameliorated by improving the accuracy of the model, and work is in hand to do this, but they clearly illustrate the sensitivity of estimates of fluxes to the sea to apparently small changes in estuarine dynamics.

Acknowledgements

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Estimation of ecological exergy using weighing parameters determined from DNA contents of organisms – a case study

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Key words: ecosystem, eutrophication, exergy, management, ecological orientor

Abstract

We studied the changes of exergy and specific exergy with data of benthic macrofauna communities, periodically sampled along an estuarine gradient of eutrophication in the Mondego estuary (Western Portugal). Exergy estimates were calculated from organism biomass, based on weighing factors for the relative content of exergy per unit of biomass determined from DNA contents of organisms. Results were discussed in terms of both the macrofauna biomass production and the structural organisation of the system. Estimates for the exergy indices provided useful indications for the evaluation of environmental impact due to the eutrophication process. Different average values for the indices of exergy and specific exergy were estimated relatively to areas with different levels of eutrophication, in the 'spatial' gradient of eutrophication. Higher exergy levels and lower exergy content per unit of biomass (specific exergy) were associated to populations more stabilized or areas less perturbed. Additionally, the index of specific exergy seemed capable of providing indications for the qualitative alterations in the communities (in temporal and spatial terms) that go in the direction of the observations made in this ecosystem.

Introduction

With regard to environmental management, efforts have been directed to the identification and description of certain ecosystem features, which are regularly changed by self-organising ecological development (Odum, 1969; May, 1974; Wilson, 1975; Odum, 1983; Okubo, 1986; Weber et al., 1989; Costanza et al., 1992; Jørgensen, 1992; Jørgensen et al., 1992; Woodley et al., 1993; Schneider & Kay, 1994a; Müller, 1997; Baird, 1998). These systems attributes can be described as emergent or collective properties which are expected to be regularly optimised during ecosystems development (Müller 1996; Wiegleb & Bröring, 1996; Bröring & Wiegleb, 1998; Bossel, 1998; Müller & Fath, 1998). This means that certain states of such attributes can be taken as ecological *orientors* (Bossel, 1992) or *attractors*, that is, stages that an ecological system usually develops towards (Bossel, 1992, 1998; Patten, 1997; Müller & Fath, 1998). Such *orientors*, described as aspects, notions, properties, or dimensions of systems, are useful criteria to describe and evaluate the system's developmental stage as ecological indicators with a more broad perspective (Bossel, 1992, 1998; Schneider & Kay 1994a, b; Müller et al., 1998; Müller & Fath, 1998; Jørgensen & Nielsen, 1998; Marques & Nielsen, 1998; Marques et al., 1998a, b; Kutsch et al., 1998). In a modelling context, the general properties indicated by the *orientors* are technically translated in terms of mathematical algorithms designated as *goal functions* (Müller & Fath, 1998; Bossel, 1998; Jørgensen & Nielsen, 1998b; Nielsen et al., 1998). This permits the development of models with a more dynamic structure, which means models with parameters (properties) that can change according to certain goal functions (Nielsen, 1990, 1992, 1995; Jørgensen, 1992c; Jørgensen & Nielsen, 1998a, b; Nielsen et al., 1998). This way, these structural dynamic models include and describe changes in species composition and trophic structure of ecosystems (Nielsen, 1994, 1995; Bastianoni & Marchenttini, 1997; Jørgensen & Bernardi, 1997;

Jørgensen & Nielsen, 1998b; Nielsen et al., 1998), resulting in an improved predictive capability and better environmental management (Jørgensen & Nielsen, 1998b; Marques et al., 1998a, b; Marques & Nielsen, 1998; Nielsen et al., 1998; Zölitz-Möller & Herrmann, 1998).

Some orientors have 'emerged' from interdisciplinary discussions embracing the fields of Thermodynamics, Succession Theory, and Network Theory (Bass, 1998; Bröring & Wiegleb, 1998; Jørgensen & Nielsen, 1998a, b; Marques et al., 1998a, b; Patten, 1998; Svirezhev, 1998; Ulanowicz, 1998). The *exergy* is a concept derived from Thermodynamics, interpreted as a function expressing energy with a built-in attribute of quality (in terms of energy potential to perform work) (Jørgensen & Mejer, 1977, 1979, 1981; Jørgensen, 1992a), a measure of the contrast between a system and its surrounding environment (Wall, 1986; Schneider & Kay, 1994b; Jørgensen & Nielsen, 1998a), or an estimate for the maximum capacity of energy to perform useful work as the system proceeds to equilibrium with its surroundings (Brzustowski & Golem, 1978; Ahern, 1980. Quoted from: Schneider & Kay, 1994a). It has been suggested as a potential indicator of ecosystems state of development and health (Nielsen, 1990; Jørgensen et al., 1995; Fuliu, 1997; Marques et al., 1997, 1998b, b; Müller, 1997). The ecosystem's ability of self-organization permits it to deal with external changes/perturbations, and in response the system may reorganize its structure and functioning (e.g. matter gradients, trophic relations, flows of energy, etc.) (Søndergaard et al., 1990; Zhou et al., 1996; Jørgensen & Padisak, 1996; Marques et al., 1998a; Jørgensen & Nielsen, 1998b). In time, the systems evolve to different states of 'contrast' relative to their surroundings, and consequently their departure from thermodynamic equilibrium will be associated to their actual states of development (Jørgensen, 1992; Schneider & Kay, 1994a, b). Therefore, it has been suggested that changes of exergy can be indicative of alterations in ecosystem structure or functioning, and be useful in methodologies for the environmental management from a enhanced point of view.

Due to the high complexity of ecosystems, direct measurements of exergy are not feasible as we cannot consider and make estimations of the properties for all the components of an ecosystem. Nevertheless, in the context of environmental management, it is acceptable to use models considering only the components of a system more relevant for solving a particular problem. This way the indirect estimation of exergy is achievable regarding the system's (model) composition. This function is computed as a global summation for the components of a system (model), where each term of the global sum takes into account the relative concentration of the corresponding component and its relative departure from a thermodynamic reference state expressed in terms of its *exergy* content (Mejer & Jørgensen, 1979; Jørgensen et al., 1995). Considering detritus (dead organic matter) as a common reference state, the departure from this reference state for each (living) component, is achieved by the combination of the probability of producing detritus and the probability associated with the component's genomic dimension in terms of its genome 'maximum coding capacity' (Jørgensen et al., 1995; Fonseca et al., 2000). Exergy may also be expressed by the ratio of exergy to the total biomass of the system, being referred to as the *specific exergy* of the system, thus expressing the exergy content per unit of biomass in the system, for a given moment.

In this study, we have obtained estimates of ecological exergy and specific exergy from biomass data of benthic macrofauna communities, periodically sampled along an estuarine gradient of eutrophication in the Mondego estuary (Western Portugal) (Marques et al., 1997; Pardal, 1998). We aimed to investigate to what extent the estimation of ecological 'exergistic' indicators can assist in the assessment of the structural organisation and functioning of the system.

Description of sites studied

The Mondego estuary consists of two arms surrounding a small island (the Murraceira island), joining at about 1 km from the sea (Fig. 1). The two arms differ in terms of hydrological, physical, chemical, and sediments characteristics: the northern arm is deeper (4–8 m during high tide, variable tide amplitude of 2–3 m) than the southern arm, which is almost completely silted up in the upstream areas (Duarte & Reis, 1993; Marques et al., 1993, 1997; Pardal 1998). Consequently the freshwater discharges occur mainly through the northern arm, while water circulation in the southern arm is dependent on the tides and the discharges of freshwater from a tributary (the Pranto river) that is controlled by a sluice. The south arm can be considered almost as a coastal lagoon system in terms of hydrological properties (Marques, 1989; Marques et al., 1993, 1997; Pardal 1998; Lillebø et al.,

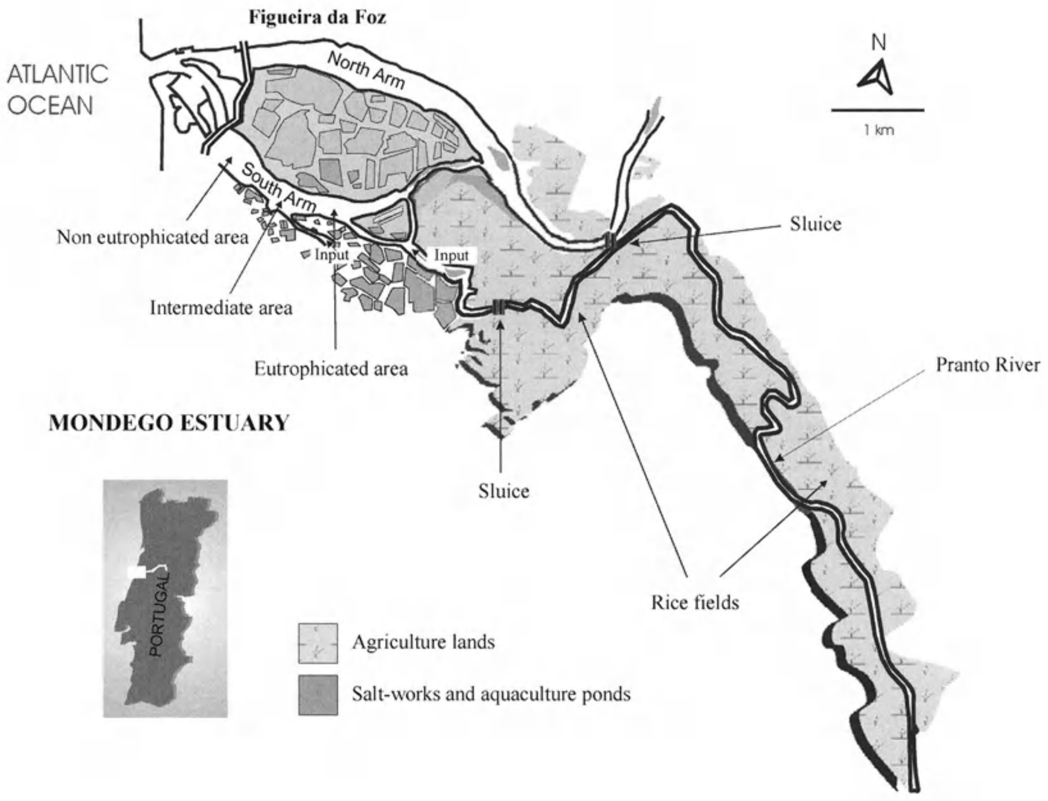

Figure 1. Representation of the Mondego estuary (Western Portugal) with the location of the sampling stations along the gradient of eutrophication.

1999; Martins et al., 2000), with a considerably higher residence period of water bodies in the inner areas of the south arm than in the north arm (Pardal, 1998).

This estuary is the localization of many harbor functions (both commercial and recreational purposes), salt-works, and ‘fish-farms’. It is also the receiver of discharges of urban and industrial sewerage systems, and chemical compounds (e.g. fertilizers and pesticides) from farmlands (mostly rice-fields) located in upstream areas (Marques et al., 1993; Pardal, 1995; Flindt et al., 1997; Azeiteiro, 1999). The drainage from farmlands represents a significant discharge of nutrients (e.g. nitrogen and phosphorous) into the water column (134 tons/year of nitrogen) (Flindt et al., 1997; Pardal, 1998). This fact and the level of confinement of the southern arm of the estuary contribute to the eutrophication of the system along a spatial gradient. As a consequence of this eutrophication process, qualitative changes have been verified in this ecosystem, where a progressive replacement of a macrophyte community, consisting of slow growing species (*Zostera*), by free floating (or partially, floating), fast growing species (e.g. *Enteromorpha* spp.) is being observed. This shift in primary producers may have consequences on the structure of communities, reflected in its specific composition, with the dominance of *r* strategists in more eutrophicated areas, and, in general, on the system’s productivity, which may result in a new trophic structure (Marques et al., 1998a; Pardal, 1998; Lillebø et al., 1999; Lopes et al., 2000; Pardal et al., 2000).

In this study, we have considered three study areas along the gradient of eutrophication. The less affected area, considered as non-eutrophied, is by the mouth of the south arm of the estuary, where a macrophyte community (*Zostera noltii*) can be found. We designate this area as the *Zoostera* meadows. On the other hand, the most affected area (eutrophied) is localised at a more upstream area, where the macrophytes have disappeared in the course of the last decade. Here green-macroalgae blooms occur, essentially of *Enteromorpha* spp., depending upon the weather conditions verified each year (Pardal, 1998; Pardal et al., 2000; Martins et al., 2000). A third study area was localised between the other two areas and regarded as an area at a stage of intermediate eutrophication (Pardal, 1998; Lillebø et al., 1999; Pardal et al., 2000).

Material and methods

Sampling and laboratory procedures

This study corresponds to a period of 13 months, from February 1993 to March 1994. During this period of time, samples of macrophytes, macroalgae, and associated macrofauna were collected, during low water tide, every 2 weeks at the three study sites described above (A – *Zoostera* meadows; B – intermediate eutrophied; and C – highly eutrophied) (Fig. 1) (Pardal, 1998; Lillebø et al., 1999; Pardal et al., 2000). Cores (10 cores per site; 143 cm of section; 15 cm depth) were placed individually in plastic bags and sieved (500 μm mesh) using estuarine water within an hour of sampling. The retained materials (sediment, macrophytes or algae, and macrofauna) were transferred to plastic bottles and preserved with 4% formalin in estuarine water. Macroinvertebrates were gathered and identified almost always to the species level. Afterwards, the correspondence into trophic collections (herbivores, filter feeders, detritus feeders, carnivores, and omnivores) was established (Pardal, 1998). Materials were dried at 60 °C for 72 h and weighed to the nearest 0.01 mg. Samples were subsequently combusted for 8 h at 450 °C and the correspondent biomass estimated and expressed as $g{\cdot}m^{-2}$ AFDW (Pardal, 1998).

Exergy estimation

Estimates of ecological exergy (Ex) and specific exergy (spEx) were calculated from the biomass of organisms ($g{\cdot}m^{-2}$ AFDW) using weighing factors for the specific exergy content of biomass for each component of the system (model). Approximate estimates of ecological exergy (Ex) were given by Jørgensen et al. (1995, 2000):

$$\mathrm{Ex} \approx \sum_{i=1}^{N} \beta_i \cdot c_i, \qquad (1)$$

where c_i is the concentration of the component i (e.g. biomass of a given taxonomic group or functional group) in the system with N components, and β_i is a 'weighing factor' expressing the specific exergy content per unit of biomass for that component. We used weighing factors (β) determined from organisms' DNA content (C-values) as described in Fonseca et al. (2000) (Table 1). In brief, the total 'genome' lengths of organisms are estimated from the DNA content in cells nuclei (their corresponding 'C-values'), and assumed as topmost limits for the 'overall coding capacity' of organisms' genomes. Then, these estimates can be 'converted' into probabilities associated to the genetic information content of organisms and used to calculate the parameters (β) to weigh the exergy content per unit of biomass from organisms. Results were expressed as g detritus exergy equivalents$\cdot m^{-2}$, since detritus (i=1) was considered as a reference state for the computation of exergy estimates. Estimates of specific exergy (spEx) were given by the ratio of ecological exergy (Ex) to the total biomass of the system ($\mathrm{Biom._{Total}}$) for each instant, according to (Jørgensen & Nielsen, 1998b):

$$\mathrm{spEx} = \mathrm{Ex}/\mathrm{Biom._{Total}}. \qquad (2)$$

Therefore, results were expressed as exergy·unit of biomass^{-1}.

Table 1. Weighing factors (β) to estimate exergy for different groups of organisms. The concentration of each organism was multiplied by the respective weighing factor to estimate the exergy content of biomass as described in Jørgensen et al. (1995). Parameters provided in Fonseca et al. (2000) were determined using nuclear DNA contents of organisms

Organisms/system compartments	Weighing factor β
'Detritus'	1
Annelids	
Polychaets	50
Arthropods	
Insects	70
Crustaceans	230
Molluscs	
Gastropods	450
Bivalves	760
Echinoderms	
Asteroideans/Echinoideans	360
Chordates	
Fish	800

Results

The values obtained for the annual biomass ($g{\cdot}m^{-2}$ AFDW) of macrofauna found in each of the three stud-

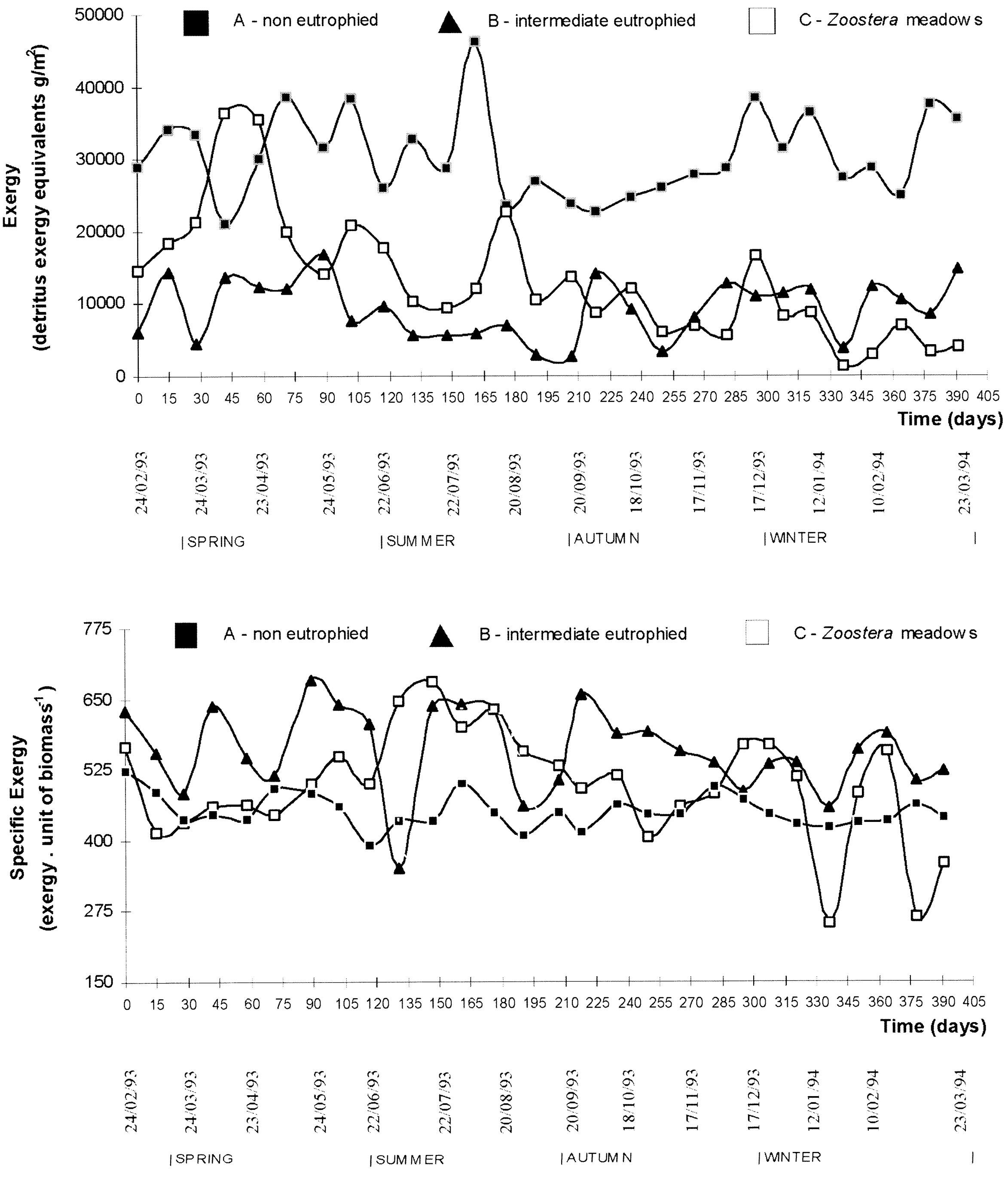

Figure 2. Variation of exergy (2.1) and specific exergy (2.2) in Zoostera meadows (A), intermediate eutrophied (B) and most eutrophied (C) areas in the Mondego estuary, for the period from to 24th of February 1993 to 23rd of March 1994. Exergy (2.1) was estimated from the macrofaunal biomass based on the use of weighing factors. The exergy content per unit of biomass was estimated from the total biomass for the period considered.

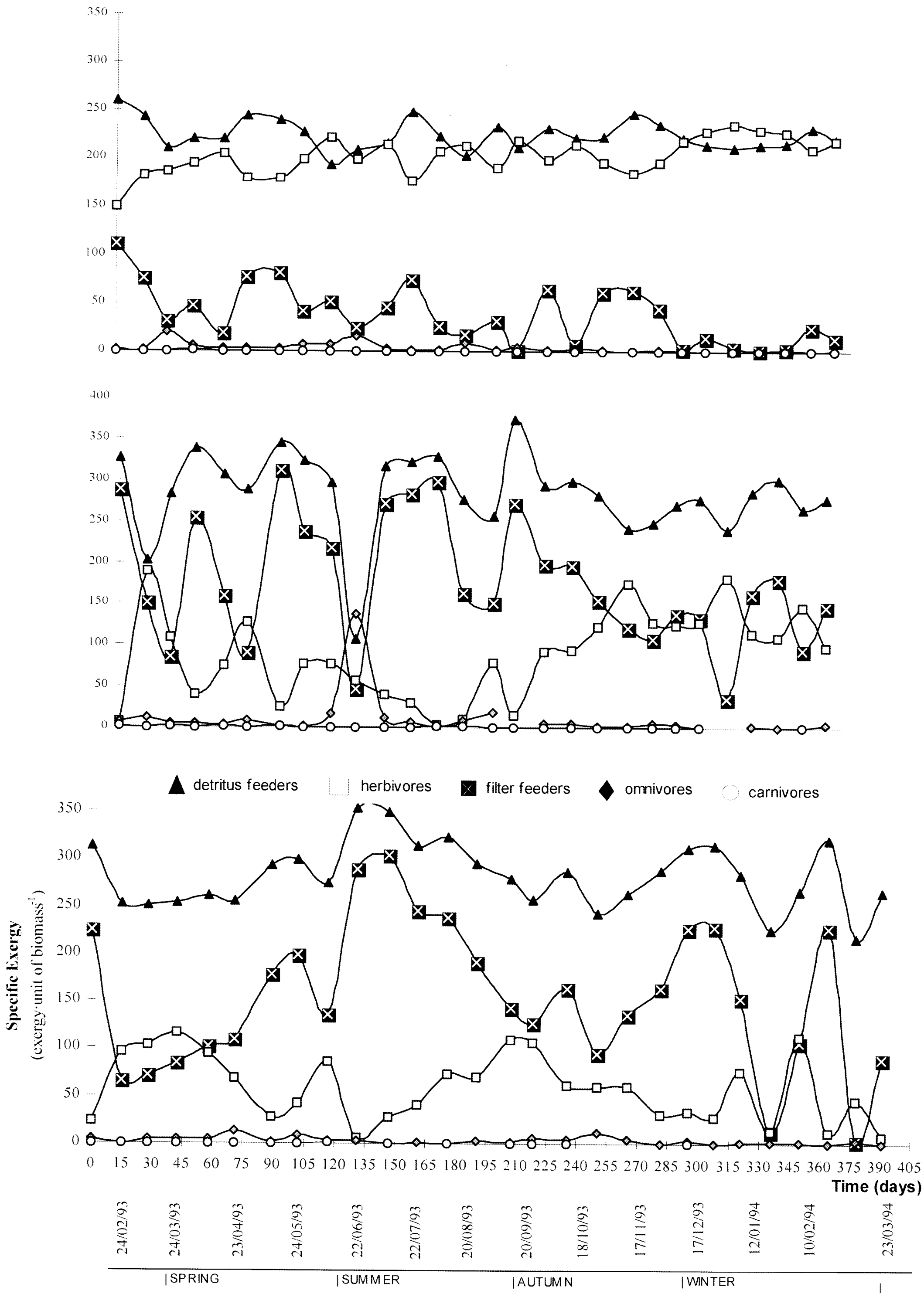

Figure 3. Variation of macrofaunal biomass in the Zoostera meadows (3.1), intermediate eutrophied (3.2) and most eutrophied (3.3) areas in the Mondego estuary, considering organisms assemblaged into trophic guilds (herbivores, filter feeders, detritus feeders, carnivores, and omnivores).

Table 2. Annual and average annual values of biomass density, exergy and specific exergy associated to the macrofauna found along the gradient of eutrophication in the Mondego estuary, for the period from to 24th of February 1993 to 23rd of March 1994

System features:		Study sites		
		A Zoostera meadows	B eutrophied	C highly eutrophied
Biomass	Annual	1893	461	743
(g·m^{-2})	Average[a]	68±12	16±7	27±19
Exergy	Annual	853 961	258 250	368 572
(g detritus exergy equivalent·m^{-2})	Average[a]	30499±5964	9223±4046	13 163±8718
Specific Exergy	Annual	12 616	15 645	13 891
(exergy·unit of biomass^{-1})	Average[a]	451±29.8	559±75.2	496±100.3

Average[a]=average annual value±standard deviation.

ied sites (A – *Zoostera* meadows; B – eutrophied; C – highly eutrophied) are given in Table 2. The estimates of exergy (detritus exergy equivalent g·m^{-2}) and specific exergy (exergy·unit of biomass^{-1}) associated with the biomass, are found in this table, also. The annual variation of biomass and the corresponding contributions associated to each trophic guild are depicted in Figures 2 and 3, respectively. The exergy content per unit of biomass (specific exergy) is represented in Figure 4, with regard to the contributions from organisms' biomass in terms of trophic groups (detritus feeders, herbivores, filter feeders, carnivores and omnivores). In Figure 5, the biomass of primary producers from the studied areas is represented: macrophytes correspond to biomass of *Zostera noltii* and macroalgae to biomass of *Ulva*, *Chaetomorpha*, and *Enteromorpha*.

The highest annual standing stock of macrofauna biomass (Table 2) was verified in the less eutrophied area (study site A – *Zoostera* meadows), corresponding to the *Zostera noltii* community. The biomass values in the most eutrophied area (study site C – highly eutrophied; macroalgae community) were higher than in the less eutrophied area (study site B – eutrophied). The same pattern was found with regard to the estimates of exergy (Table 2). On the contrary, the annual average estimate of structural exergy for the study site B (559±75.2; units: exergy·unit of biomass^{-1}) was higher than the average estimates determined for the other two areas (A: 451±29.8; C: 496±100.3; units: exergyþunit of biomass^{-1}). With regard to the specific exergy landscapes as depicted in Figure 4, the specific exergy contributions of the several trophic guilds in the area B seem to oscillate between the levels found in the areas A and C. Moreover, the exergy content per unit of biomass of detritus feeder and filter feeder organisms are higher in the eutrophied areas, especially for the filter feeder organisms during late spring and early summer (Fig. 4).

As shown in Figure 5, during spring and early summer, the *Enteromorpha* bloom contributed to the high levels of macroalgae in the most eutrophied area (study site C), but was followed by the macroalgae crash, corresponding to a drastic reduction of the total biomass. In area B the densities of macroalgae were lower than in the most eutrophied area (site C), but its levels were maintained longer in time, until early autumn.

Discussion and final remarks

Ecosystems may be considered as complex systems resulting from "biotic, physical, and chemical components of nature acting together as a non-equilibrium dissipative process" (Schneider & Kay, 1994a). Like the living organisms, ecosystems use high quality energy as 'fuel' in metabolic processes of matter and energy conversion, enabling them to maintain their structure or increase its internal order (Schrödinger, 1944; Jørgensen et al., 1999). The energy quality can be understood in terms of work potential of the system relatively to a state of equilibrium (Wark, 1995), its exergy content. High quality energy means a higher exergy content and, on the other hand, low quality energy implies an energy of lower potential to perform

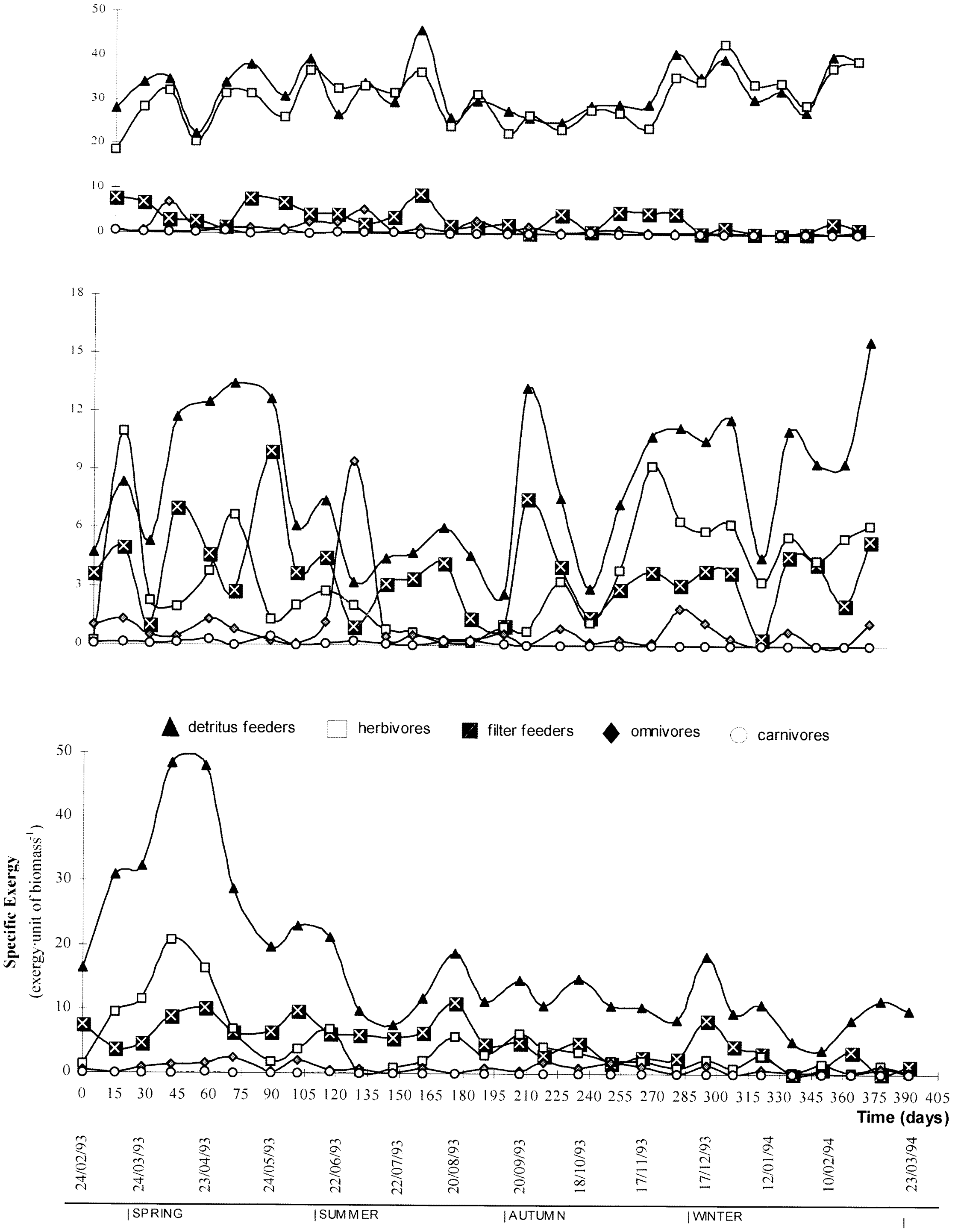

Figure 4. Variation of specific exergy in Zoostera meadows (4.1), intermediate eutrophied (4.2) and most eutrophied (4.3) areas in the Mondego estuary, considering contributions from organism biomass of each trophic guild.

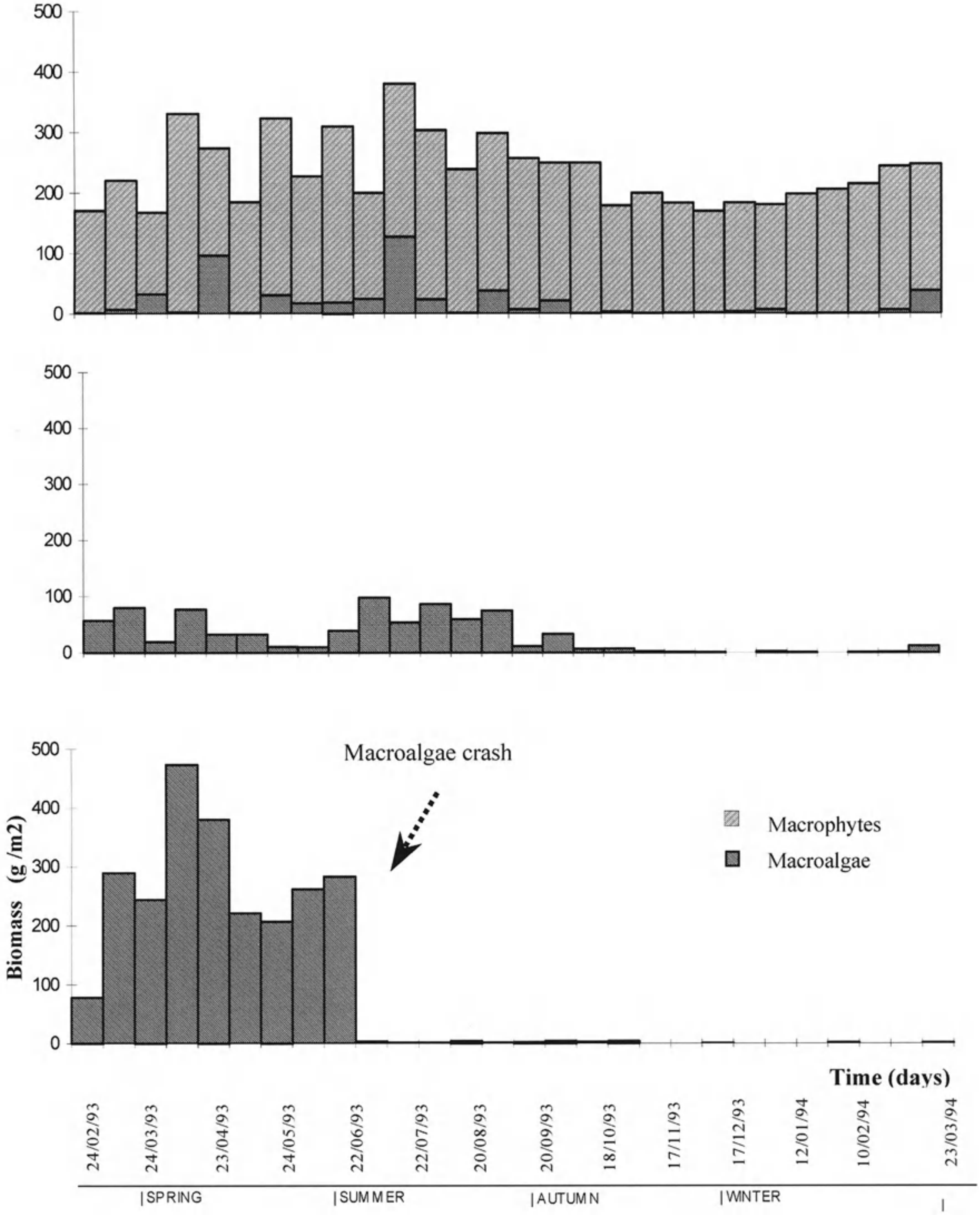

Figure 5. Variation of biomass density of primary producers found in Zoostera meadows (5.1), intermediate eutrophied (5.2) and most eutrophied (5.3) areas in the Mondego estuary. Macrophytes correspond to *Zostera noltii* biomass and Macroalgae to *Ulva*, *Chaetomorpha*, and *Enteromorpha* biomass. Data provided in Pardal (1998).

work, which is dissipated as heat without a temperature gradient (Straþkraba et al., 1999; Jørgensen et al., 1999). In ecological terms, the concept of exergy is interpreted as a measure for the available energy invested by an ecosystem in maintaining and building its structure far from the thermodynamic equilibrium (Jørgensen, 1992b, c; Schneider & Kay, 1994a, b, 1995; Jørgensen, 1997; Jørgensen et al., 1999). The use of this concept in ecological studies, although necessarily with great approximations, requires the estimation of the relative amount of exergy embedded in the biomass. Therefore, the weighing parameter β is taken as a discriminator of the exergy (organizational) level of biomass in Equation (1). We have used values for the β parameter described in Fonseca et al. (2000), which were determined from the nuclear DNA contents of organisms as more operational approach for the estimation of exergy from organism biomass in Fonseca et al. (2000).

Values of exergy and specific exergy were calculated from the biomass of the different organisms periodically sampled along an estuarine gradient of eutrophication in the Mondego estuary. During the first 4 months of the sampling period, the biomass of primary producers was followed by higher exergy estimates in the system, particularly in the most eutrophied area. Also, exergy estimates in the most eutrophied area were, on average, higher than in the intermediate eutrophied area. Therefore, it seems that higher exergy levels correspond to more stabilized levels at either end of the eutrophication gradient, as found in the communities around both the *Zostera* meadows (non-eutrophied area) and *Enteromorpha* (most eutrophied area) populations. On the other hand, on average, higher specific exergy estimates were associated with the populations in the intermediate eutrophied area, and in the most eutrophied area after the macroalgae crash. According to the results, the macroalgae crash may be interpreted as a disturbance bringing the most eutrophied area to the same state as the intermediate eutrophied area. Thus, higher exergy levels and lower exergy content per unit of biomass (specific exergy) seem to be associated with more stabilized populations or less perturbed areas. This way, these indices may provide different and complementary indications about the structure/functioning of the system. Additionally, from one viewpoint, the specific exergy contributions of the several trophic groups in the area B seem to oscillate between the levels found in the areas A and C. On the other hand, the exergy content per unit of biomass of detritus feeder and filter feeder organisms are higher in the eutrophied areas, particularly for the filter feeder organisms during late spring and early summer. This is a putative indication of the shift from a primary production based situation towards a detritus based food web (Marques et al., 1997). Therefore, the results correspond to field observations describing that in the intermediate eutrophied area the recruitment of new individuals (juveniles) may occur both from the macrophytes and the macroalgae communities, but the levels of disturbance do not stimulate the establishment of new organisms (Pardal, 1998).

Results obtained from the estimation of both exergy and specific exergy, for the considered estuarine system, were based on weighing factors (β) determined by the application of organisms' nuclear DNA contents (Fonseca et al., 2000). Results provided good indications of the qualitative alterations occurring in the system and are useful in the evaluation of the environmental impact due to the eutrophication process.

From a more global point of view, what might be the contribution of this study for the general hypothesis on exergy optimisation? It appears more and more evident that a critical need in Ecology is to develop theoretical scaffolding capable of explaining results obtained up to now in terms of an accepted pattern. The aim should be to build in Ecology the equivalent of physical theory – 'equivalent' in the sense that the laws explaining observations derive from a very few fundamental laws (Jørgensen & Marques, 2001). The efforts of many researchers over quite a few years will be necessary to develop such a theoretical network, and progress will be stepwise and, considering the difficulties, slow (Jørgensen & Marques, 2001). Exergy optimisation in developing ecosystems represents a node in this theoretical network.

It is evidently critical to find ways to integrate large sets of observations and databases, and to interpret the scientific basis, results, and predictions clearly. In the present paper we intended to contribute to establish a bridge between empirical observations and theory. In fact, why should empirical researchers, environmental managers, and lay stakeholders care at all about theory? The only possible answer is, because a theoretical frame provides the context wherein researchers and others can interpret and integrate empirical results. Without integration and interpretation of raw observations within a consistent theoretical frame, only description is possible, not basic understanding that can be transmitted.

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Trophic status based on nutrient concentration scales and primary producers community of tropical coastal lagoons influenced by groundwater discharges

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Key words: coastal lagoon, eutrophication, primary productivity, phytoplanktonic biomass, residence time, groundwater, nutrients

Abstract

Changes in the rate of primary production as an indication of trophic status of aquatic ecosystems have been one of the major indicators of their health. Despite different approaches been devised for evaluating this process, few are useful for comparison and generalization of their results. The coastal zone ecosystems of Yucatan (SE Mexico) exhibit hydrological variability closely associated with the groundwater discharges, which are the only freshwater sources in the coastal ecosystems. In order to learn about the trophic status of karstic tropical coastal lagoons, three of them (Celestun, Chelem and Dzilam) located in Yucatan (SE, Mexico) were monitored for inorganic nutrients, Chl-*a*, phytoplankton and macrophyte productivity. The nitrate concentrations suggest that the trophic status was influenced by fresh water springs, being meso-eutrophic in Celestun, oligotrophic in Chelem, and mesotrophic in Dzilam. In the case of ammonium ions, the three lagoons were mesotrophic, indicating that processes such as remineralization play an important role in the trophic dynamic of these shallow ecosystems. According to phosphate concentrations, Celestun and Dzilam were mesotrophic, and Chelem was oligotrophic. External inputs of phosphate and bioturbation by waterfowl may be responsible of these differences. Primary productivity at Celestun was greater than at Chelem and Dzilam lagoons and the contribution by seagrasses was significant in all three. It is found that indices based on nutrient concentration and phytoplankton biomass are useful as an indication of trophic status in groundwater influenced coastal lagoons. Moreover, estimations on the total system productivity, and the relative contribution of each primary producer, is a holistic approach useful for understanding trophic dynamic in shallow tropical coastal ecosystems.

Introduction

The coastal lagoons of Yucatan are depressions roughly parallel to the coast, separated from the sea by a biogenic sand bar known as a 'barrier island'. Sediment deposition through wave action and coastal currents contributes to the formation of these physiographic features (Lankford, 1977).

A distinct aspect of the Yucatan Peninsula is the lack of rivers and a well-developed network of underground water flowing towards the open sea carrying with it inorganic nutrients such as NO_3 and SRSi, and showing a hydrological variability strongly associated with the discharge from fractures in the freatic mantle as springs (Herrera-Silveira & Comin, 1995). It amounts to 90% of all freshwater available at the coast and has been recognized by its role as nutrient source.

As the metabolism of non-conservative materials in coastal lagoons is closely tied to ecosystem trophic condition, it is of general interest to study the trophic status and the factors related to it in different coastal ecosystems.

The existing balance between the input of inorganic nutrients from different sources and their use or transport through assimilation, sedimentation or export from the system is important in evaluating coastal trophic state. All trophic classifications are based on arbitrary divisions of a continuum, and in marine ecosystems, no specific criteria have been yet established as it is the case in freshwater ecosystems (Havens et al., 1999).

In Yucatan Peninsula, there are three coastal lagoons in its north and northwestern portion: Celestun, Chelem and Dzilam, which though sharing geological, geomorphological and climatological characteristics, they exhibit significant functional differences due to their physical behavior, including fresh water/sea water balance, which in turn affect water residence time and physical and chemical water column gradients; primary productivity and total organic production. Moreover, they show different degree of human apropiation, been Dzilam the most pristine system.

The variety of ecological scenarios of the coastal lagoons of the Yucatan Peninsula, provides the opportunity to try different approaches to learn about their trophic status. This knowledge is basic for sustainable management of these ecosystems, which are under rapid development.

Study area

Celestun

Celestun lagoon is a shallow body of water, 1.5 m average depth, located at the northwest extreme of the Yucatan Peninsula (20° 45′ N, 90° 15′ W; Fig. 1). It is surrounded by well-developed mangrove vegetation, connected to the sea through a 410 m wide mouth and shows a strong salinity gradient, with 5–15 psu in the lagoon interior and 33–38 psu at its mouth due to fresh groundwater discharge from springs (Herrera-Silveira, 1994).

Water flow has been modified by the construction of a bridge crossing its central portion. Intense tourism activity also stresses the submerged aquatic vegetation which is significant to the ecology of a wide variety of fauna, including the flamingo (*Phoenicopterus ruber ruber*) and commercially important species.

Chelem

Running parallel to the northern coastline of the Peninsula is the Chelem lagoon (21° N, 91° W; Fig. 1), also a shallow one, 1.4 m average depth, measuring 14.7 km long by 1.8 km maximum width, with an overall surface area of 15 km^2. It is connected to the sea by a 225 m wide artificial opening which serves as a harbor with access to the Port of Yucalpeten. The north shore is the area of largest population density on the coast of the State of Yucatan, including the towns of Chuburna, Chelem, Yucalpeten and Progreso. The State's largest port, which harbors 90% of the fishing fleet, is also located there. The main fresh water input comes from precipitation and from a few springs. Most of them have been separated from the main body of water by road construction, giving rise in salinity (Valdes & Real, 1994). Present restoration programs show a recovery of the submerged aquatic vegetation (Herrera-Silveira et al., 2000).

Dzilam

The Dzilam lagoon is located on the north central portion of the Yucatan coast (21° 26′ N, 88° 42′ W; see Fig. 1), and like the others is a long, shallow (1.2 m average depth) body of water running parallel to the coast and surrounded by mangrove vegetation. Of the three lagoons in this study, this one has experienced the least amount of impact, as access is only possible from the sea. Its hydrology is clearly linked to groundwater discharge, as shown by salinity as low as 4 psu. Aquatic vegetation coverage is extensive and dominated by *Halodule wrightii* and *Ruppia maritima* (Herrera-Silveira et al., 1998).

Materials and methods

Samples were taken monthly in each lagoon between August 1997 to September 1999. Ten stations were placed along their length and data collected for physical and chemical parameters (temperature, salinity, dissolved oxygen and light extinction), inorganic dissolved nutrients ($NH4^+$, NO_2^-, NO_3^-, SRSi, SRP), phytoplankton (Chl-*a*, and productivity), and macrophytes (biomass and net productivity).

The trophic status was estimated using the Carlson index based on Chl-*a* concentration (Equation (1)), as the presence of chlorophyll-*a* in the water column is considered an almost immediate response to nutrient variability (Carlson, 1977). This allows monitoring and quantification data to be used as trustworthy indicators of phytoplankton biomass variations and ecosystem trophic state:

$$\mathrm{TSI\ (Chl\text{-}}a) = 10\left[6 - \frac{2.04 - 0.68 \ln \mathrm{CHl\text{-}}a}{\ln 2}\right]. \quad (1)$$

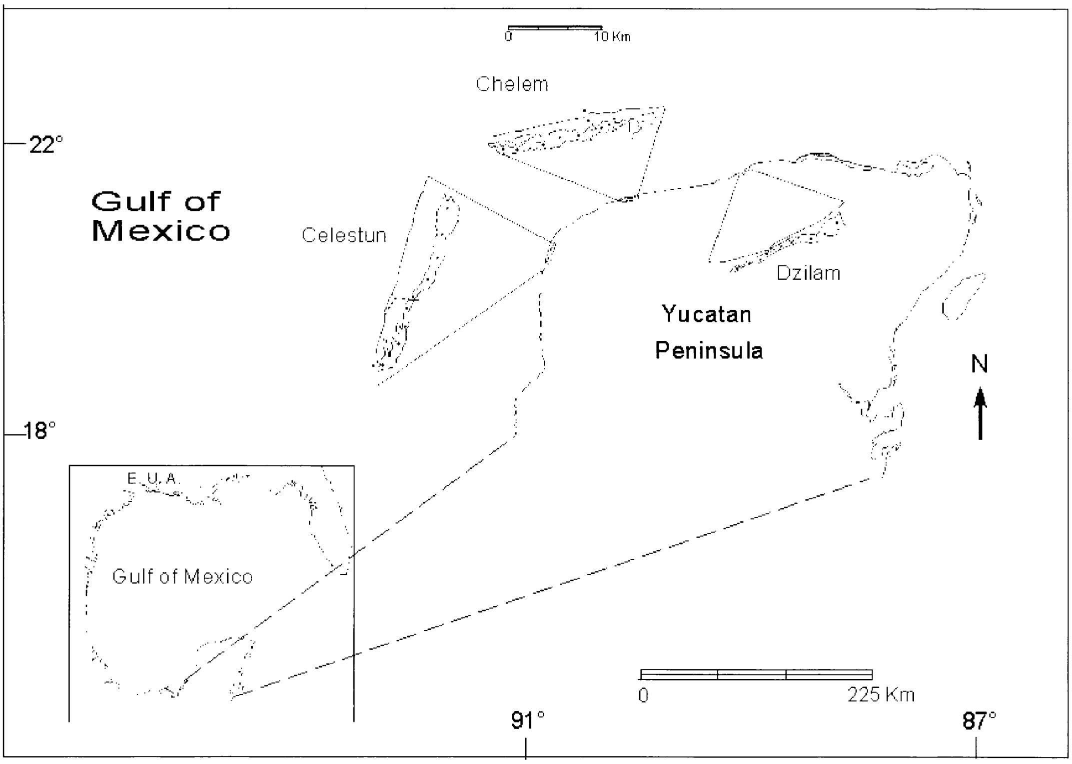

Figure 1. Coastal Lagoons of the Yucatan Peninsula.

The index was constructed using the range of possible values for Secchi disk transparency. On the basis of the relationship between algal biomass and Secchi disk transparency (expressed by the equation for vertical extinction of light in water), trophic states for the index are defined using each doubling of algal biomass (or halving Secchi disk tranparency) as the criterion for the division between each state.

The regression of Secchi disk against chlorophyll-*a* showed a non-linear element in the relationship, which necessitated a log-log transformation of the data (expressed in the ln 2 of the Equation (1)).

With the same end in mind, Karydis et al. (1983) proposed a simple empirical formula devised to be used as an index for direct determination of the nitrogen and phosphorus eutrophication levels (Equation (2)). The index tests the relationship between the total amount of a certain nutrient at a given station and its total annual amount in the surveyed area. Then, corresponding hierarchization is based on estimation done within a system, and trophic state can be evaluated for each sampling station.

Logarithmic transformations of the nutrient concentrations were used in order to normalize the nutrient values and smooth on their minor random fluctuations.

$$I - \frac{C}{C - \log x} + \log A. \tag{2}$$

The index was formulated as a ratio to provide dimensionless values. On the Equation, I is the trophic index, C the log of the total nutrient load, x the nutrient total concentration in certain station and A is the number of stations. Log A has been added to make the formula independent of the number of stations.

The scale and hierarchization produced from the Carlson and Karydis indices, specifically for this study, are: oligotrophic=1; oligo-mesotrophic=1.5; mesotrophic=2; meso-eutrophic=2.5; eutrophic=3.

A simple alternative for nutrient balance study in these ecosystems is the approach defined by LOICZ (Gordon et al., 1996). In this, three elemental terms are identified for the water balance calculation: residual volume (V_R); mixing flow (V_X); and water residence time (τ). The calculation uses salt as an indicator, because it is a conservative material, and its interchange with the sea is the most significant process effecting the water balance (Smith et al., 1997). Using this method, a box model is created that allows estimation of the water volume entering the system through connection with the adjacent marine zone, which in turn is associated with the seasonal dynamics between lagoon water interchange and atmospheric processes.

It is also possible to estimate residence time for water within the lagoon, which then gives several internal processes. Analysis of nitrogen and phosphorous loss and gain within the lagoon is based on concentration of these elements at the sources, including the residual flow determined through water balance, and within the system. This indicates if the system is dominated by an autotrophic or heterotrophic condition, as oxidation-reduction processes in sediments and the water column mediate net nitrogen and phosphorous gain and loss in a system.

Phytoplankton productivity was estimated by measuring oxygen change in light and dark, 300 ml DBO bottles incubated in duplicate for 4 h. Chlorophyll-*a* determination was carried out by passing 140 ml of water from each station through 0.45 μm millipore filters, and then extracting the pigments in the laboratory using 90% acetone. The spectrophotometry was according to the methods and equations of Jeffrey & Humphry (1975).

A PVC core sampler was used to take samples of submerged aquatic vegetation (SAV), which then were cleaned and preserved in 10% formaldehyde. In the laboratory, these samples were first identified and then dried at 70 °C for 24 h., cleaned and weighed. In an ideal case, macrophyte biomass can be considered as the remnant net growth from respiration, thus, a change in biomass during a known time interval is a measure of net productivity. Given this, macrophyte production is calculated by adding the positive biomass changes for each species at each site, and assuming that carbon content is 37% of dry weight (Westlake, 1974).

Results and discussion

The limestone platform of the Yucatan Peninsula and the physical framework that surrounds it define a large variety of environments. Processes arise in these that have special physico–chemical characteristics, and that deeply influence the hydrology and variability of coastal zone ecosystems.

Quantitative assessments of trophic state in aquatic ecosystems have been so far mainly based on phytoplankton response to the trophic nature of the environment and expressed in the form of indicators (Ignatiades et al., 1992). However, shallow coastal environments support high densities of benthic plants and periphyton, and as a result, display low phytoplankton biomass and high transparency due to competition for nutrients with the benthic producers. So submerged aquatic vegetation should play an important role in the total primary production as in the nutrient dynamics; hence, different approaches are required to establish the trophic state of a specific system, which should include the primary producers community, nutrient scales and the metabolism of the system.

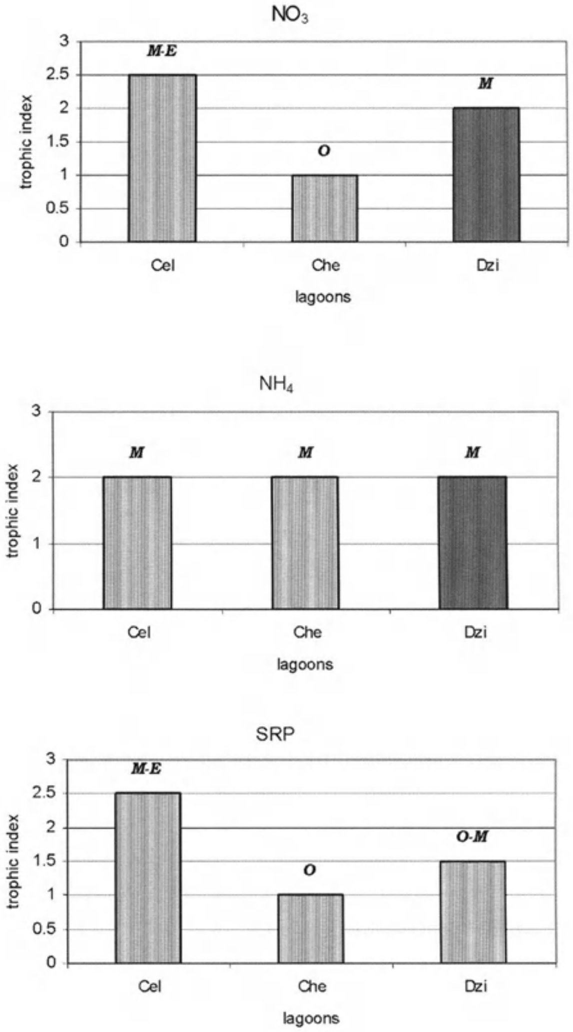

Figure 2. Trophic index related to nutrient concentration (nitrate, ammonium and phosphorus).

Through analysis of NO_3 trophic state index, the three lagoons were distinct, varying from meso-eutrophic condition at Celestun, to oligotrophic at Chelem, to mesotrophic at Dzilam (see Fig. 2). Nitrate

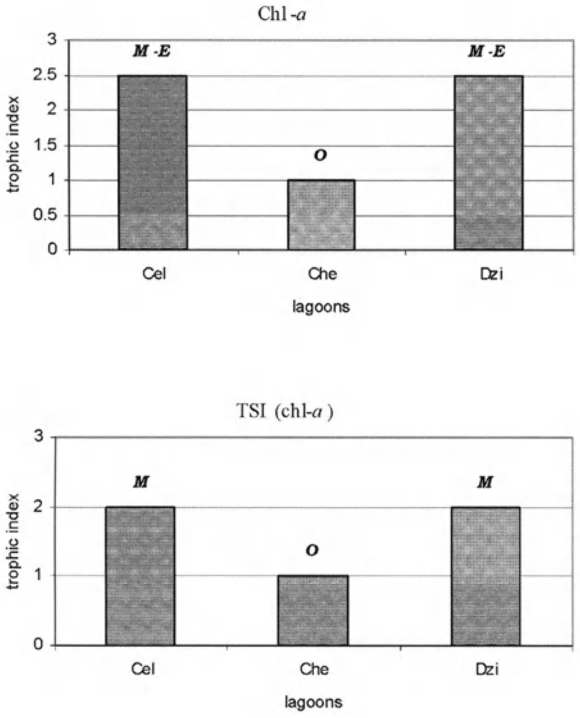

Figure 3. Trophic state index related to Chlorophyll-*a* concentration: (a) Carlson trophic state index; (b) Karydis index.

has identified as the most sensitivity nitrogen specie in characterizing pollution levels for coastal marine ecosystems (Karydis & Tsirtsis, 1996).

The variation in the trophic level for each coastal lagoon on this study may be associated with the influence of NO_3 input from groundwater sources. The intensity of this input is different in each system due to differential fresh water discharge related with the permeability of the soil. It is greater in the Celestun (western) and Dzilam (eastern) areas, than in the Chelem (northern) area. The greater groundwater discharge at Celestun and Dzilam results in a preponderance of NO_3 in the metabolism of these systems. The lower input at Chelem, due to less fresh water discharge from springs, results in lower NO_3 values and an oligotrophic state.

The mesotrophic condition found in the three lagoons through NH_4 analysis, shows that the reduced nitrogen input from neighboring subsystems or from autochthonous organic matter decomposition processes is moderated by assimilation in productive processes and exportation from the lagoon (see Fig. 3). The near eutrophic state of the Celestun lagoon calls attention to the importance of hydrodynamic conditions as water residence time as a factor which could modify the trophic status of the coastal ecosystem (Boyer et al., 1997; Arhonditsis et al., 2000). Good interchange and flow of water in coastal lagoons and bays should avoid extreme effects of the eutrophic condition as the hypoxia with severe consequences on the global metabolism of the system.

In the case of the phosphorous index, the condition for Celestun was mesotrophic tending towards eutrophic, for Chelem it was oligotrophic, and for Dzilam it was mesotrophic (Fig. 4). The trophic level at Celestun may be associated with the ornithofauna of the lagoon. The direct phosphorous input from aquatic birds excrement, and the indirect input from the bioturbation, could cause in the sediments a significant source of these nutrients in this ecosystem, and rapidly resuspended by the shallowness of these type of systems (Bieng et al., 1988; Herrera-Silveira, 1994), and favoring the assimilation by the primary producer community.

The difference between trophic state defined by phosphorus and Chl-*a* in Dzilam lagoon could reflect the relative larger water residence time in this system compared to the other lagoons assessed; as a consequence, phytoplankton is able to assimilate nutrients (nitrogen also) in a level that diminishes phosphorus concentration and records a minor value for this index than Chl-*a* index.

Both the Celestun and Dzilam lagoons were meso-eutrophic for the Chl-*a* index, which is consistent with the trophic state index (TSI, Carlson). The water residence time (τ) in both these lagoons is greater than that for Chelem, which leads to an increase in phytoplankton biomass as a consequence of longer periods spent in high nutrient waters (Fig. 4). This may also favor greater phytoplankton productivity in the Celestun and Dzilam lagoons in comparison to the Chelem one.

The phytoplankton biomass accumulation is consequence of physical and chemical factors, as water flow and nutrient inputs (Harris, 1986) and establishes not only variations on nutrient concentration but influence seagrasses development by light attenuation and nutrients competition.

It should be mentioned that the TSI results for Chelem do not reflect the ecosystem conditions. This may be due to these index was established considering to phytoplankton as the most important primary producer (Carlson, 1977), however, in these shallow coastal lagoons the submerged aquatic vegetation is most important. On the other hand, the short water residence time, which lessens the tendency towards eu-

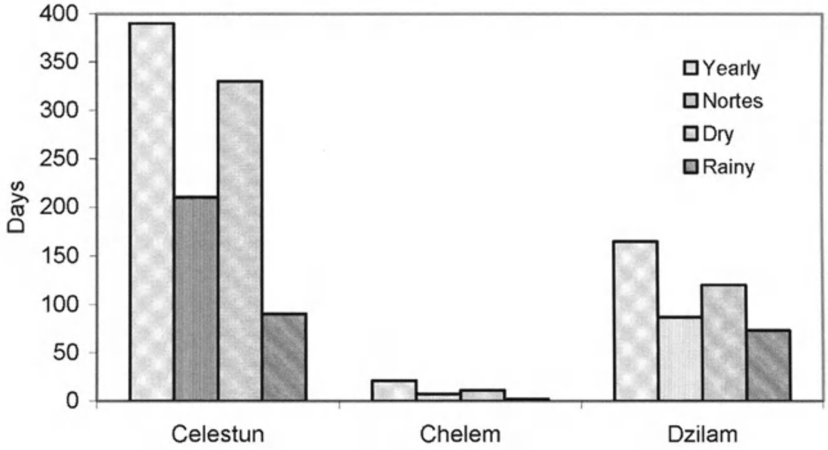

Figure 4. Water residence time in three coastal lagoons of Yucatan, Mexico during an annual cycle.

trophication may lead to underestimation of its actual trophic state when based on these indices.

The total primary productivity values were greater in Celestun than in Dzilam and Chelem (see Fig. 5), though the relative contribution of grasses, phytoplankton and macroalgae to production was similar between Celestun and Chelem (Fig. 6). For Dzilam, the contribution of macroalgae to production was the lowest, with phytoplankton and seagrasses maintaining a more complementary condition in the organic production process.

One of the initial symptoms of eutrophication that has been proposed for coastal lagoons is an increase in macroalgae biomass and coverage, and the substitution of the seagrasses community with macroalgae (Stevenson et al., 1993). In this sense, when Dzilam is compared with the others, it shows a production congruent with its low human impact, and may be taken as an ecosystem in a high state of conservation, which could be used for comparative studies.

The indices used in this study have also been employed in freshwater and coastal ecosystems, and in scenarios differing from the karstic ecosystems as those found in different parts of the world as Australia, Florida, Greece, North-Spain and Yucatan (Buddemeier, 1996).

In the coastal lagoons of the Yucatan Peninsula, reactive phosphorous availability is subject to variations in redox potential and pH promoted by inorganic particle adsorption and phosphorous coprecipitation. Also, the nitrogen cycle is notably influenced by spring discharge, which contributes a significant amount of nitrogen in the form of NO_3 (Herrera-Silveira, 1994). However, nitrification is also increased by the presence of organic matter, which can favor oxidation-reduction processes in sediments as has been observed in Chelem Lagoon (Valdes & Real, 1994).

Given these conditions, the use of indices based on nutrient concentrations or phytoplankton biomass can be useful in defining the trophic status in tropical karstic coastal lagoons and the complete evaluation of the system state could be improved determining the total system production, through the estimation of the relative contribution of each component to this production, as a holistic approach towards understanding the trophic condition of these coastal ecosystems.

Conclusions

The Celestun lagoon exhibits a tendency towards the eutrophication, especially in terms of inorganic nitrogen. This emphasizes the importance of preserving low water residence times in order to keep this condition stable.

The oligotrophic condition of the Chelem lagoon, as seen by nutrient concentration, may be associated with the short water residence time. This leads to an underestimation of the actual trophic state of this lagoon, which experiences heavy anthropogenic pressure.

According to the trophic state index (TSI), Dzilam and Celestun have similar mesotrophic conditions. However, water residence time in Dzilam is considerably greater, and may cause temporary nutrient accumulation in the inner part of the system (useful

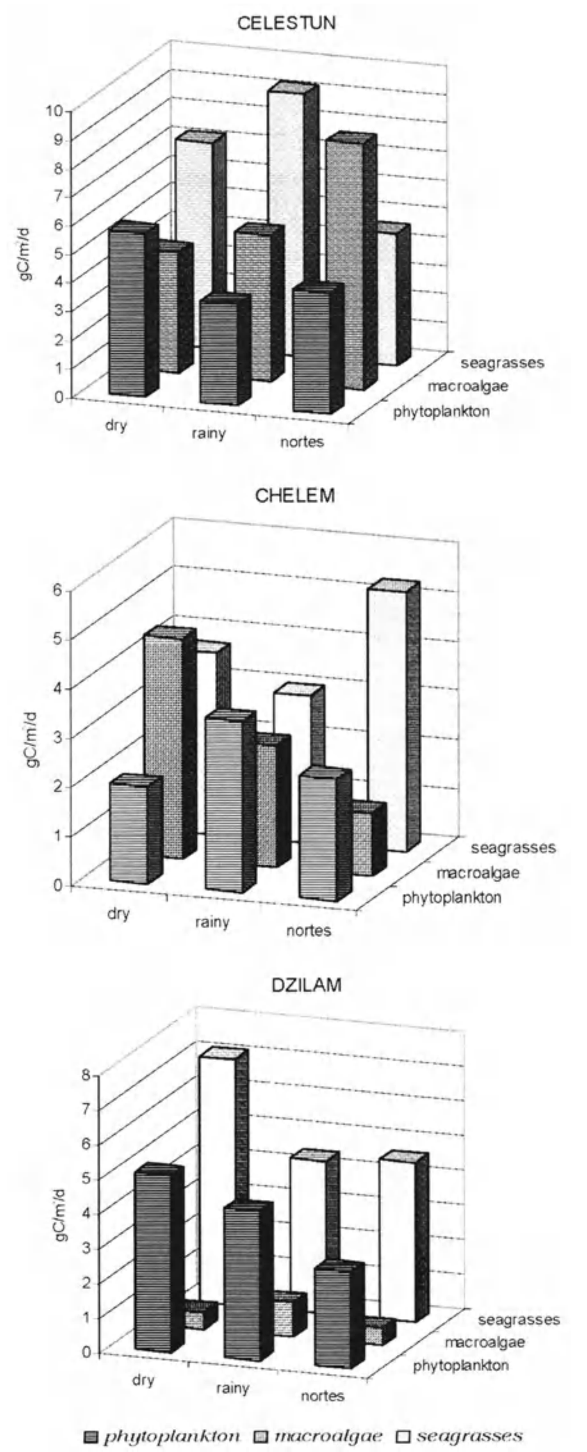

Figure 5. Annual and seasonal primary productivity in three coastal lagoons.

for organic production), and lead to a more balanced condition between autotrophic and heterotrophic.

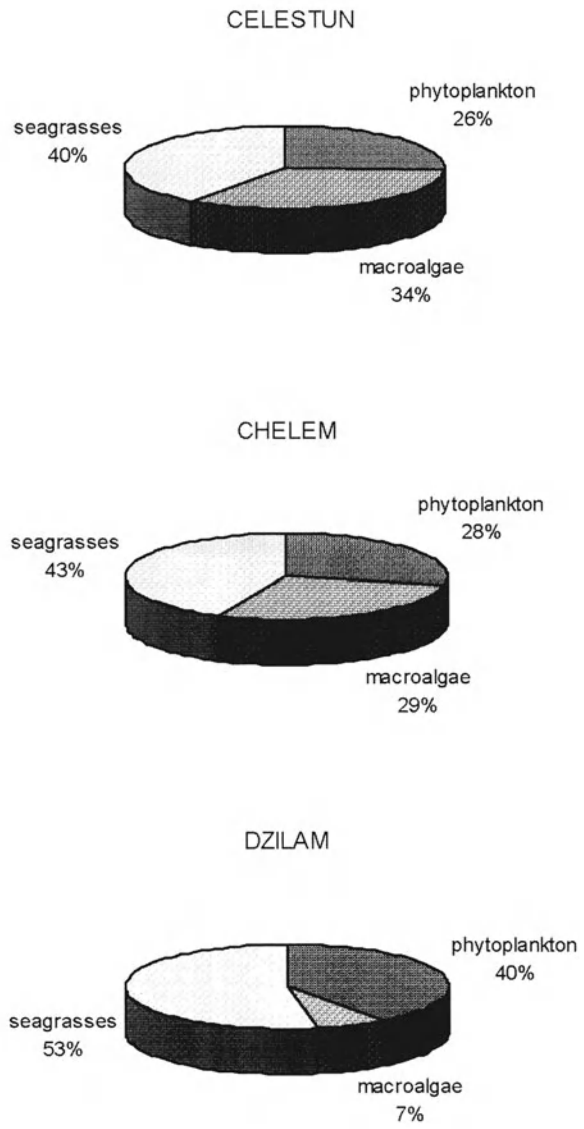

Figure 6. Relative contribution of seagrass, macroalgae and phytoplankton to total primary productivity in three coastal lagoons of Yucatan Peninsula.

The high seagrass production and coverage in the Dzilam lagoon are congruent with its meso-eutrophitrophic natural state and high level of system conservation.

The contribution of each component to total primary productivity provided a holistic approach to understanding trophic state, whereas those based on nutrient concentrations are useful in defining the processes responsible for the observed trophic level.

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'Ferry-Boxes' and data stations for improved monitoring and resolution of eutrophication-related processes: application in Southampton Water UK, a temperate latitude hypernutrified estuary

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Key words: chlorophyll-fluorescence, phytoplankton blooms, Ferry-Box, *in situ* monitoring, hypernutrification

Abstract

To provide detailed observations of algal bloom development in Southampton Water which is a hypernutrified, macro-tidal estuary (mean tidal range 3.2 m, low suspended load <100 g m^{-3}), a ferry running between Southampton and Cowes on the Isle of Wight, was fitted with an instrument package ('Ferry-Box'). Measurements were made of temperature, conductivity, turbidity, and chlorophyll-fluorescence at a data rate of 1Hz. For comparison a data station which measured the same variables was operated at a fixed site in the estuary. In 1999 the Ferry-Box achieved reliable operation with a data return over 95%, for the fixed data station the return was 92%. From this data spatial and temporal variations in chlorophyll *a* concentrations have been mapped. The maps show the development of blooms in different areas of the estuary, through the spring and summer, in relation to tidal and weather conditions. In 1999 conditions were such that the spring bloom increased in intensity through a spring tide (maximum chlorophyll *a* 55 mg m^{-3}), which coincided with calm weather with high light levels (irradiance). This was followed by a sequence of seven blooms, the development of which can be related to changes in the tidal energy, irradiance and nutrient supply.

Introduction

Increased inputs of nutrients (nitrate and phosphate) from rivers and the atmosphere to the seas of the European shelf are well-documented (Howarth et al., 1996). On the south coast of the UK, Southampton Water receives high inputs of nitrate from the rivers Test and Itchen ($>$400 mmol m^{-3}) and direct discharges into the estuary of phosphate from sewage works are also significant, equivalent to a freshwater input concentration of $>$20 mmol m^{-3} (Xiong, 2000). At the temperate latitude of the UK the limited amount of light energy or irradiance available in autumn and winter imposes an annual cycle of growth and decay on plankton populations (Tett, 1990). The development of blooms in estuaries depends on the existence of appropriate physical conditions (Sinclair et al., 1981; Cloern, 1996). In Southampton Water concentrations of chlorophyll *a* of up to 40 mg m^{-3} have been reported (Purdie, 1996; Crawford et al., 1997). In this system, as the supply of nutrients from land is relatively constant through the year, the main spring bloom tends to be followed by a sequence of blooms through the spring and summer (Kifle & Purdie, 1993). The intensity of bloom development is known to vary markedly at different positions in the estuary at different times in spring and summer (Kifle & Purdie, 1993). Although previous observations based on a limited number of boat surveys had identified the repeated occurrence of blooms, the amount of sampling was insufficient to precisely define when blooms were occurring relative to changing conditions in the estuary. To do this requires the collection of data at high temporal resolution, which in turn requires continuous observations using *in situ* instrumentation. Previously an offshore data buoy has been used in Southampton Water (Wright et al., 1997). This system had disadvantages in that it only provided data at one position in the estuary and was relatively expensive to maintain. Maintenance was expensive because it could only be

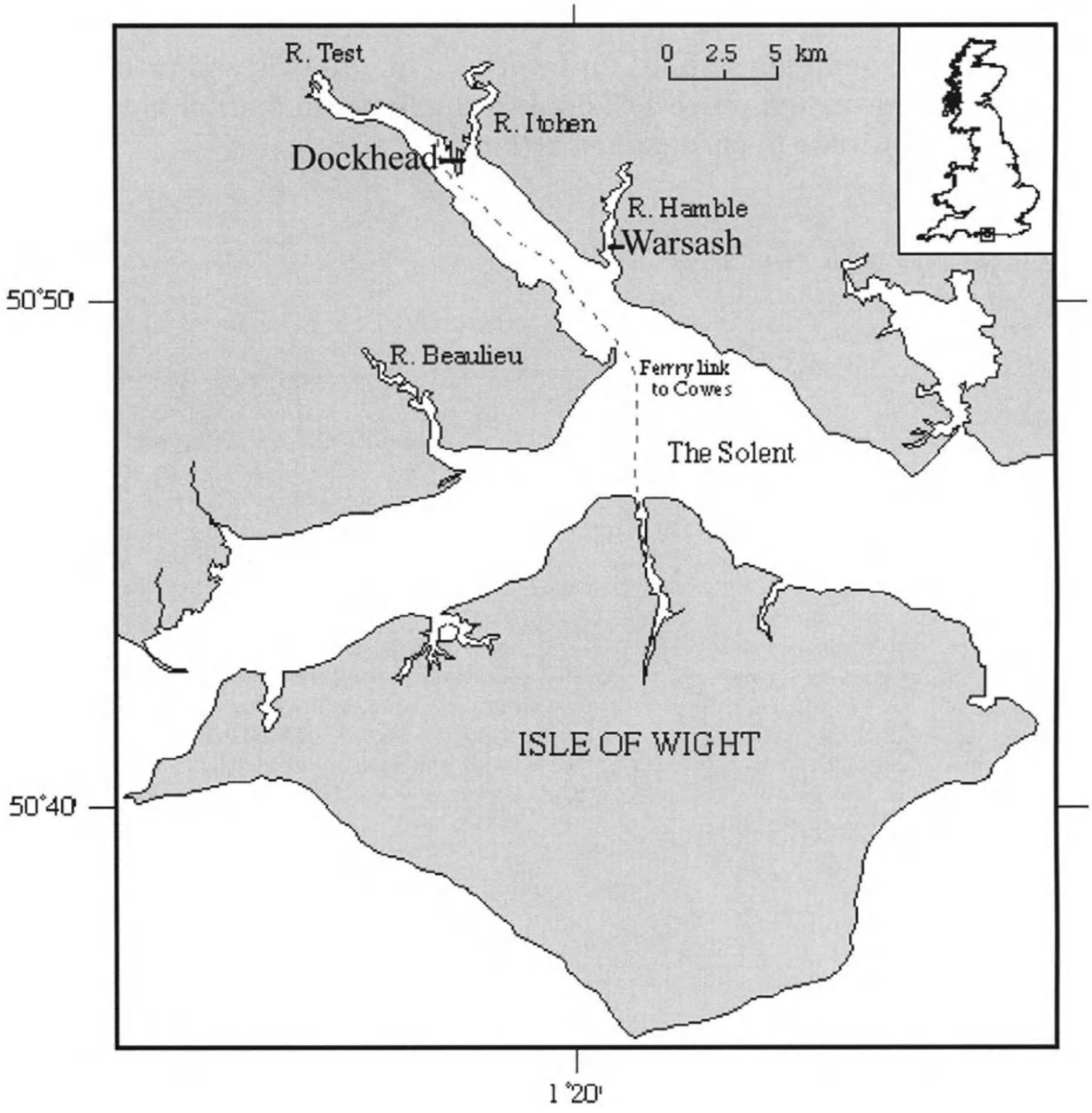

Figure 1. Map of Southampton Water showing the position of the Dockhead data station and the route of the Red Falcon Ferry.

serviced from a boat and because a minimum of three people were required to carry out the work.

To address the problem of a lack of spatial coverage and to gain easier access to the instrumentation than was the case with the data buoy, two new more easily accessible systems were installed to make measurements in Southampton Water in 1999. A set of sensors at a fixed position and a similar set of sensors carried repeatedly along the length of the estuary on a ferry. The instruments fixed at a permanent location against the dock wall in Southampton Water, the 'Dockhead' sensors, are at a point close to the confluence of the Test and Itchen arms of the estuary (shown on the map in Figure 1). The other similar set of sensors were fitted as a 'Ferry-Box' (Tziavos & Flemming, 1998) on the Red Funnel Ferries' ferry the 'Red Falcon'. It operates between Southampton, Town Quay along the length of Southampton Water, across the Solent, to Cowes on the Isle of Wight (see Figure 1). The instrumentation on the 'Red Falcon' delivers up to 16 surveys a day. Measurements are made of chlorophyll-fluorescence, turbidity, salinity, temperature, ships position and time, at a rate of 1 Hz, which is equivalent to a spatial resolution of about 7 metres when the ferry is at full speed. The 'Ferry-Box' concept – installing instrumentation on ferries running regular routes to collect systematic oceanographic data – has been strongly promoted by groups planning a Global Oceanographic Observing System GOOS (Prandle & Flemming, 1998; Tziavos & Flemming, 1998). So far, reported applications of such systems have tended to be in relatively open seas – the Sea of Japan (Harashima et al., 1997), the North Sea (Swertz, et al., 1999) and the Baltic Sea (Leppanen, 2000). A test of the idea on a shorter route is described here.

In this paper we examine the practical application of techniques for long term *in situ* data collection to see how well a record of bloom events can be captured in a hypernutrified estuary known to be affected by recurring blooms in spring and summer. The equipment used is described. The quantity and quality of data obtained during the first two seasons of operation from March to September 1999 is assessed. The data are interpreted along side other available information to test the assumption that continuous observations do provide good definition of when blooms occur relative to changing physical conditions in an estuary.

Equipment and methods

Dockhead system

The instruments in use at Dockhead are based on a W.S. Ocean Systems Ltd Coastal Monitor (CLM-2) system. It includes an *in situ* sonde with attached sensors, which contains the electronics, controls the data sampling routines and records the data from the sensors. The specifications of the sensors are listed in Table 1. The *in situ* sonde is located in a robust plastic tube attached to the dock wall 1 m above chart datum. This method has the advantage that the sonde can be easily recovered by hand from the dockside without the assistance of divers. Power is supplied from a single 12-V lead acid battery, which is trickle charged by a wind generator mounted on the 3-m high mast. The battery, junction box for the sensor cables and the data transmission equipment are contained in a protective housing bolted to the dockside stone work. The variables measured at this monitor are water temperature, conductivity (used with water temperature to calculate salinity), turbidity and fluorescence. The data obtained are recorded on the internal memory of the sonde. Measurements are made at 10-min intervals and telemetered ashore using a Vodaphone-Paknet cellular radio system.

'Red Falcon' Ferry-Box

The Ferry-Box was commissioned on the Red Falcon in March 1999. A sensor package was installed in the engine room of the ship. The measurement specifications of the standard sensors on the Ferry-Box are the same as at Dockhead and are described in Table 1. Water feeding the sensors is taken from the ship's engine cooling water supply between the intake and the ship's heat exchanger. The Ferry-Box does not require a pump as the pressure drop between the take off and return points provides sufficient flow through the system. Wiring connects the sensors in the engine room to a computer housed on the ship's bridge. The computer is in turn connected to the Paknet data transceiver and to a GPS decoder box which has an antenna mounted on the roof of the bridge. Power supply is from an un-interruptable power supply unit housed on the bridge along side the computer. The sensors measure fluorescence, salinity, pressure, water temperature and turbidity, global positioning sensor (GPS) data is also recorded to relate variables measured to the position of the ferry in the estuary. The data obtained are recorded on the hard disk of the PC and stored as raw data files (1 Hz) and as 1-min averages of the raw data. So that the performance of the system can be monitored remotely, samples of the data are transmitted from the Ferry-Box to a receiver ashore via a Vodaphone-Paknet cellular radio system. A single reading of the data, averaged over 1 min, is transmitted every 15 min. At approximately weekly intervals the ship is visited, the equipment checked and data downloaded from the PC.

Boat survey

On the 2 August 1999 a survey was carried out using a research boat along the track of the ferry. During the boat survey, water was collected from an intake at 1 m depth and pumped to a bucket on the deck. Sub-samples were taken at regular positions along the length of the estuary (at the sites shown in Figure 1). The salinity of the samples was measured using

Table 1. Specifications for the Dockhead and Ferry-Box sensors

Parameter	Sensor type	Range	Accuracy	Resolution	Response
Fluorescence	LED	0.03–100 mg m^{-3} (Dockhead); 0.03–3% 30 μg/l Ferry-Box	± 0.03 or	0.01	–
Water temperature	Thermistor	−2–32°C	0.01°C	0.0005	2s
Conductivity	Inductive	0–65 mS/cm	0.01 mS/ cm	0.001 mS/ cm	120 ms

a WTW salinometer. Two 50-ml sub-samples were filtered through 25-mm diameter Whatman GF/F filters and the filters stored immediately in a freezer for chlorophyll *a* analysis. Chlorophyll *a* was analysed ashore using a fluorometer (Parsons et al., 1984) and was calibrated against a chlorophyll *a* standard, the concentration of which was measured using a spectrophotometer. The survey results were used to evaluate the Ferry-Box data calibration.

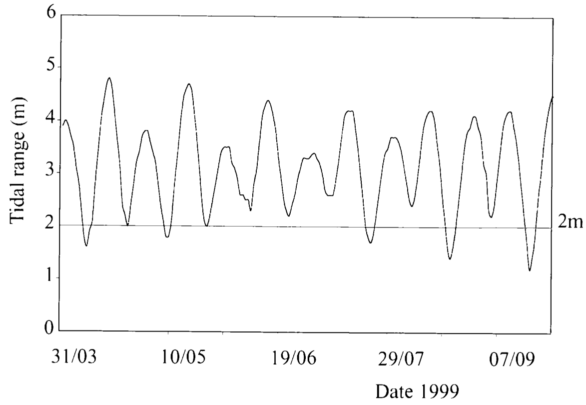

Figure 2. The predicted daily tidal range in Southampton Water from April through September 1999 (Admiralty, 1999).

Calibration methods: Dockhead And Ferry-Box

The amount of chlorophyll *a* present in seawater is used as an indication of plankton biomass. For the Dockhead and Ferry-Box, the concentration of chlorophyll *a* was calculated from the output of the fluorometers. The estimates of *in situ* chlorophyll *a* concentrations for the Dockhead fluorometer were calculated after deriving a calibration equation, made from laboratory measurements of extracted chlorophyll *a* concentrations from samples collected close to the sensor compared to the contemporaneous data from the sensor. The Dockhead sonde was hauled from the water at weekly intervals and cleaned along with the attached sensors to remove any biofouling. To this end a 0.05-m^{-3} barrel was filled with surface water collected by bucket hauls. The sonde was then placed in the barrel for 20 min (to ensure two readings from the sensors). The surface water was then sampled for the laboratory determination of salinity and chlorophyll *a*. Potential problems can occur with this method of sub-sampling, as plankton may settle out in the container (Gardner, 1977) so the sub-samples were well mixed. The relationship of the sensor readings to the chlorophyll *a* concentrations may vary with different species present but an overall relationship for the year, with a correlation coefficient, r^2 of 0.78 (n=24), was applied to the Dockhead data and gave consistent results.

Discrete samples are not taken on the ferry, as the water flow in the engine room cannot presently be tapped for regular calibration. Up until 12 July the Ferry-Box chlorophyll *a* was determined by comparison with the Dockhead data as the ferry crossed the same latitude (50.88°N) as the Dockhead site. The Ferry-Box sensor was prone to fouling on a fairly regular basis, due to the ship being laid up every three nights. After cleaning the sensors on 12 July bio-fouling was not seen again and the signal was consistent from this date. After 12 July the Ferry-Box fluorometer was calibrated using known quantities of chlorophyll *a*. The Fluorometers were immersed in a range of suitably diluted solutions of chlorophyll *a* standard in laboratory conditions and the output measured to calculate the calibration equation for conversion of fluorescence to chlorophyll *a* readings.

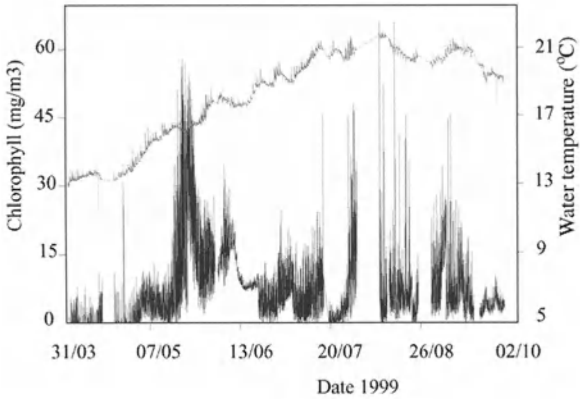

Figure 3. In situ chlorophyll-fluorescence data from the Dockhead site and the Red Funnel Ferry-Box from April through September 1999, and the corresponding water temperature data at the Dockhead site.

Results

Two important drivers of plankton blooms in estuaries are the physical stability of the water (vertical stratification) and the light energy input (irradiance). In a sheltered environment like Southampton Water physical stability is largely determined by the spring neap tidal cycle, shown in Figure 2 as the predicted sequence of daily tidal heights in Southampton Water in 1999. Changes in weather patterns are less predictable and more variable. Figure 3 shows the water temperature data, along with the chlorophyll *a* data, collected at the Dockhead site between 10 March and 17 September 1999. Major hiatuses in the Dockhead record are present in both the fluorescence and temperature traces, these are gaps in the record when the Dockhead equipment was out of service. It should be noted that the bandwidth of the Dockhead temperatures is relatively narrow and changes little through the year. The temperature variation is in the order of 1 °C, which implies some stratification, but generally this indicates that tidal stirring is too great for a significant summer thermocline to develop, thus Southampton Water is relatively well mixed with respect to temperature. Wright et al. (1997) found that the water temperature record from the SONUS data buoy provided a clear integrated signal of changes between periods of good and bad weather. Poor weather conditions correspond to a plateau, or decrease, in the otherwise increasing water temperature readings throughout the spring and summer months of 1999. These can be compared to the weather record for the region (Eden, 1999). The following poor weather periods can be identified from 1 to 12 June (when there were storms in the region), 18 to 23 June, the 26 June to 1 July and from 19 to 22 of July.

Comparison of output from Dockhead and Ferry-Box fluorometers

The chlorophyll *a* data collected from the Dockhead during 1999 are plotted in Figure 3, this figure shows the seasonal variation in chlorophyll *a* from April to the end of September. During spring and early summer the fluorometer windows tended to collect a mucous layer. Sharp drops in the Dockhead record occurred when the mucous was removed when the fluorometer was cleaned, for example on 25 June. However, these jumps are smaller (<4 mg m^{-3}) than those occurring due to bloom events (>10 mg m^{-3}) and the build-up of mucous ceased towards the end of July. Overall the percent numerical data returns for the Dockhead and Ferry-Box were 92 and 95%, respectively. The larger degree of bio-fouling of the Ferry-Box fluorometer due to less frequent service visits reduces the amount of usable data from that sensor to 85%.

The large daily range of the fluorescence data from the Dockhead site in Figure 3 is a result of the non-uniform nature of the distribution of chlorophyll *a* in the water column. The Dockhead sensor is mounted at a fixed position relative to the bed of the estuary so that as the tide goes in and out water is moving relative to the sensor in all directions. Consequently, waters of lowest salinity, which are closest to the surface of the estuary, are sampled at low tide. A simplified picture of the variation in chlorophyll-fluorescence at the Dockhead site is given by the box and whisker plot in Figure 4 which shows the daily mean and standard deviation. A series of peaks occurred through the spring and summer and the highest chlorophyll *a* levels occurred around 20 May. Sequences of small peaks follow the main peak in May and occur in the records until the end of August.

To examine if similar patterns in variation were detected by both systems, Ferry-Box data collected at the same latitude as the Dockhead data station (50.88°N) were compared with data from the Dockhead site. One week was selected (19 to 28 July 1999), a period over which a bloom developed, and the results are shown in Figure 5. The two systems sampled water at different depths and positions. The Dockhead system was at a fixed depth and sampled water throughout the water column due to the movement of the tides. The Ferry-Box always sampled surface water and the

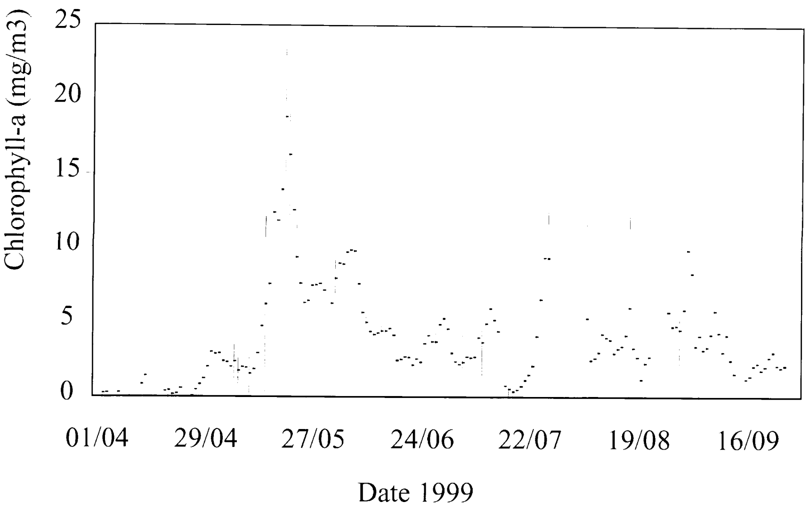

Figure 4. Variation in chlorophyll-fluorescence at Dockhead presented as a box and whisker plot showing the data for each day as the mean and standard deviation.

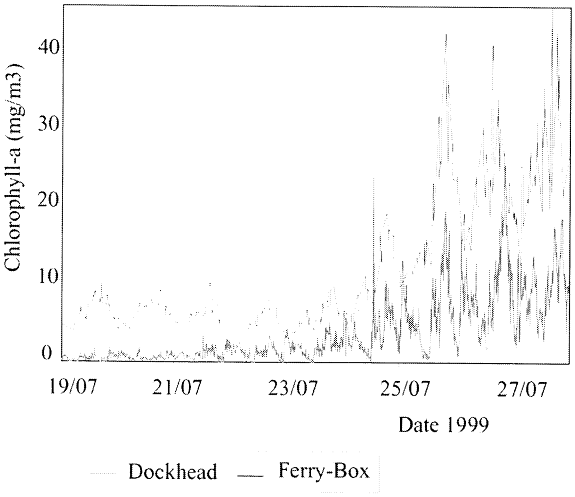

Figure 5. A comparison of the Dockhead and Ferry-Box (extracted at the same latitude as the Dockhead) chlorophyll *a* data over 8 days from 19 July 1999.

closest approach to the Dockhead was about 300 m. Also the Ferry-Box data were calibrated using the laboratory calibration and the Dockhead data using the continuous calibration. The Ferry-Box fluorometer was calibrated by relating the *in vitro* fluorescence, from chlorophyll *a* standards to *in vivo* fluorescence. This calibration procedure is not optimal. However when chlorophyll *a* measurements made from water samples collected during the boat-based survey were compared to the Ferry-Box estimates of chlorophyll *a*, from the crossing closest in time to that of the boat survey (Fig. 6), a good agreement is observed. The actual levels of chlorophyll *a* estimated from both systems are different. Discrepancies highlight a previously observed pattern namely that the absolute measurement of chlorophyll *a* concentrations *in situ* is difficult (Jeffrey et al., 1997). Despite generally lower chlorophyll *a* readings at the Dockhead site (Fig. 5) both data sets show the same patterns of increase in chlorophyll *a* during July, with similar daily cycles in the signal. This good qualitative agreement identifying the timing of periods of higher plankton growth and accumulation was true throughout the period of observations.

Patterns of blooms in Southampton Water in 1999

Little change in chlorophyll-fluorescence was seen in April (Fig. 3). In late April to early May there was an increase in chlorophyll *a*, seen as a small peak in the Dockhead trace around 2 May (Fig. 4). This was the peak day of a weak spring tide (Fig. 2). After this day the temperature record shows a change in the weather and the concentration of chlorophyll *a* declined at Dockhead. The main spring bloom developed later in May. It is seen as a distinct peak in the Dockhead data, which reached a maximum concentration of chlorophyll *a* of 55 mg m^{-3} on 17 May (Fig. 3). The corresponding daily mean value was 18 mg m^{-3} (Fig. 4). This bloom was sustained over the spring tide, peaking two days after the maximum tidal range (Fig. 2). The next clear peak in the Dockhead fluorescence record (Fig. 4) occurs around 5 June. The maximum of this bloom occurred 4 days after the

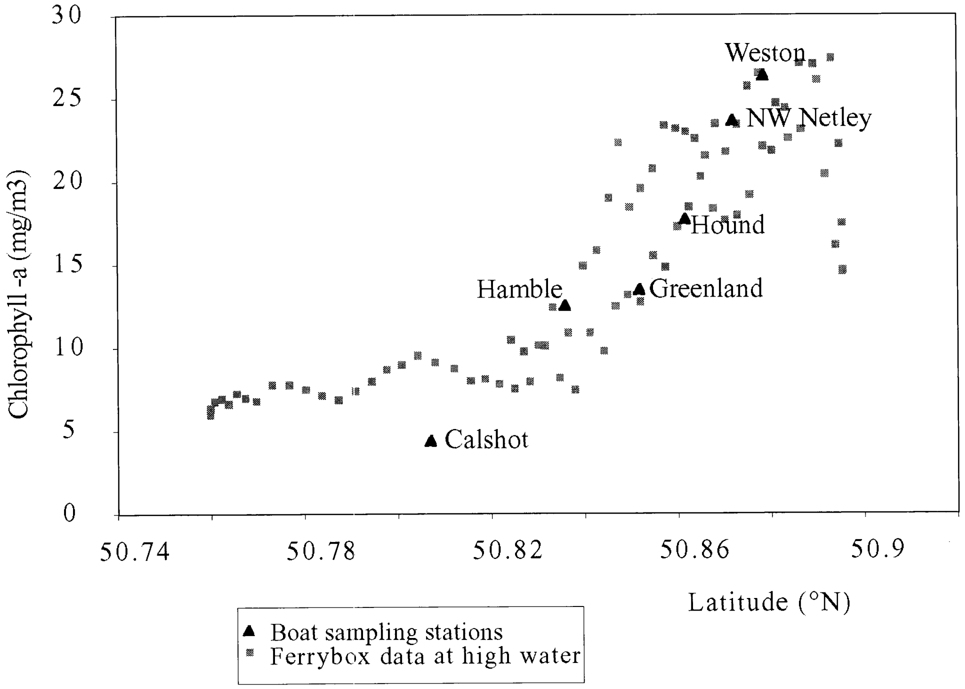

Figure 6. Comparison of the laboratory calibrated Ferry-Box data with the research boat discrete chlorophyll *a* data from standard sampling sites in Southampton Water on 2 August 1999.

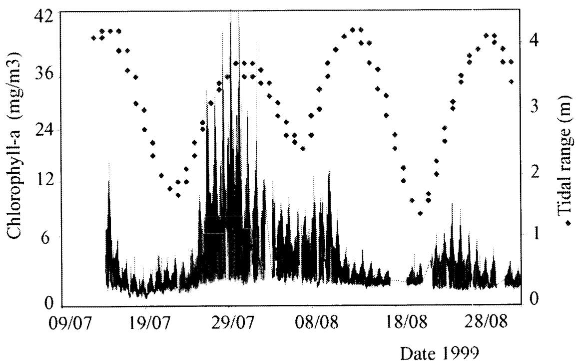

Figure 7. Detailed plot of the *in situ* chlorophyll-fluorescence data from the Red Funnel Ferry-Box from 14 July to 28 August 1999, and the corresponding data for the daily tidal range.

weak spring tide on 1 June. The bloom peaked during a period of decreasing tidal dispersion. This may be connected to the deterioration of the weather at this time. Thunderstorms and torrential rain marked this period on 2 June after which it was unsettled and cool between 3 to 7 June (Fig. 3). This may have inhibited fuller development of the bloom.

Peaks are present in the data following neap tides on 22 June and 7 July (Fig. 4). At these times peak concentrations of chlorophyll *a* in excess of 8 mg m^{-3} were measured. These peaks are consistent with the idea of blooms being dispersed by increasing tidal energy moving from neap to spring tidal conditions. A stronger bloom with chlorophyll *a* values over 20 mg m^{-3} developed around the neap tide on 22 July. On 28 July an electronic problem developed in the Dockhead sonde. The sonde had to be removed for repair so data were lost up to 6 August so that the peak values of this bloom were not recorded at the Dockhead site. The Ferry-Box data show that this bloom peaked around 27 July (Fig. 7), and chlorophyll *a* levels remained high through to the spring tide on 30 July. This was a period of clear sunny high summer weather. Subsequently, the Ferry-Box data show a succession of peaks from late July through to September with clear but successively smaller peaks in the chlorophyll *a* fluorescence signal following each neap tide (Fig. 7).

The Ferry-Box data sets from the individual ferry crossings have been merged and used to create 'contour maps' showing the change in signal against position in the estuary and the time of day. Four such daily 'maps' are presented in Figure 8. These show the distribution of chlorophyll *a* on the days of the peak chlorophyll *a* concentration in April, May, June and July 1999.

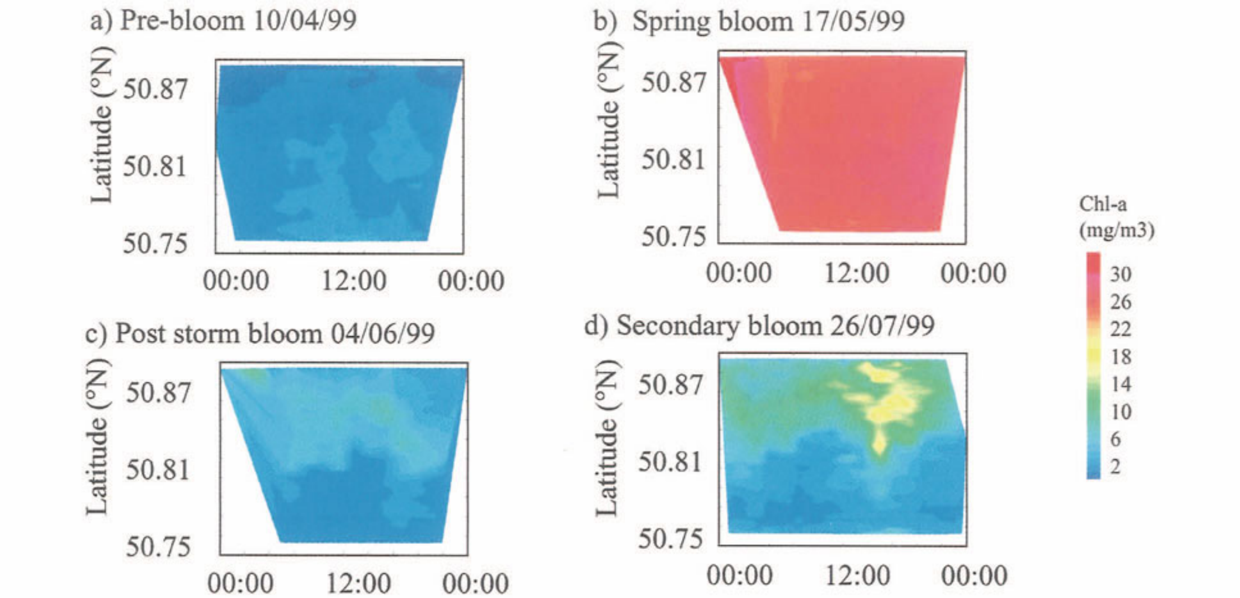

Figure 8. Contour plots of chlorophyll *a* distribution from the Ferry-Box data, at peak chlorophyll *a* concentrations, during April, May, June and July 1999.

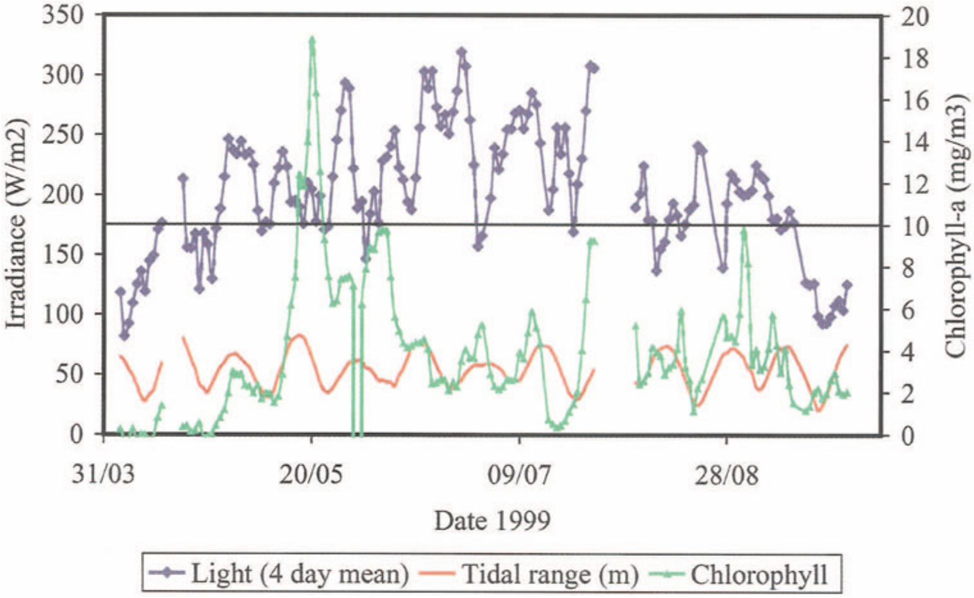

Figure 9. Light data at Everton, Hampshire (BADC, 1999), tidal height and Dockhead chlorophyll *a* data.

Discussion

The range of processes and factors, which control phytoplankton blooms in estuaries, are well documented. At the latitude of Southampton Water the annual variation of irradiance is a major control on production and limits it to the spring and summer months in all but shallow, clear water. During months when there is sufficient light many estuaries are characterised by a marked variability in phytoplankton biomass and production over periods of days or weeks. The generally accepted idea is that physical processes regulate blooms over short-time scales and short term variations in biomass are associated with short-term fluctuations in the degree of density stratification in the water column (Sinclair et al., 1981). In shallow coastal environments tidal stress tends to dominate so blooms develop on the weak tidal energy of the neap tide when dispersion is reduced and vertical stratification is more pronounced. Conversely blooms dissipate on the spring tides when the tidal energy is stronger. The balance of phytoplankton production and loss is considered to be sensitive to the rate of vertical (and horizontal) mixing in the water column (Cloern, 1996). Increased vertical mixing lowers concentrations of chlorophyll *a* due to decreased residence time in the photic zone. Reduced stratification from increased mixing on the spring tide is paralleled by a reduced phytoplankton standing crop. The effect of vertical mixing can be indirect, as it will affect the light levels received by cells; intense mixing may cause fluctuations in the light levels, which occur too fast for a cellular response. Tidal stirring also affects bottom sediments and the concentration of suspended solids in the water column, which in turn changes light levels.

The two most important controls on bloom development are available light and tidal energy. We now examine how phytoplankton blooms in Southampton are related to a critical light level and/or tidal energy state. As no measurements of light were made as part of this project irradiance data at a nearby meteorological observing site (Everton, Hampshire UK, British Atmospheric Data Centre, 1999) was used. We do not have the necessary information to estimate likely variation in *in situ* light climate in terms of photo-synthetically available radiation (PAR, Tett, 1990), however the daily global irradiance data from Everton is plotted in Figure 9. Blooms generally occur in Southampton Water between May and September (Kifle & Purdie, 1993; Iriarte & Purdie, 1994). Looking at these dates in Figure 9 suggests a critical total global radiation value at this latitude of the order of 200 W m^{-2} day^{-1}. This is close to the critical light level of 193 W m^{-2} day^{-1} which is drawn on Figure 9 (estimated by Sinclair et al., 1981, after Riley, 1957). For preliminary analysis of the Dockhead and FerryBox data we use the figure of 193 W m^{-2} day^{-1} as an indicator of those periods when reduced light levels may or may not have limited phytoplankton growth.

To find a similar guide number for tidal range we have considered the paper of Monbet (1992). From a study of 40 estuaries world-wide, Monbet (1992) showed that despite similar nutrient levels macrotidal estuaries showed significantly lower chlorophyll *a* than micro-tidal estuaries (mean tidal range <2 m). Consequently systems with smaller tidal ranges tend to be more sensitive to dissolved nutrient concentrations (Monbet, 1992). Exceptions occurred in rivers with high suspended particulate materials, high freshwater flow and discharge of toxic substances. Southampton Water has a high tidal range of up to >4.5 m (mean 3.2 m). It should be classed as a macrotidal system in which dispersion would be expected to limit the occurrence of blooms. But the variation in tidal range is large from <2 to >4.5 m, and it is also an estuary with a relatively low suspended sediment concentration (<100 g m^{-3}, Xiong, 2000). Therefore, at times it might be expected to show the characteristics of a micro-tidal estuary. On this basis we first look at those occasions in the tidal spectrum (Fig. 2) where the neap tidal range was less than 2 m. These should be the times most suited to bloom development. For the effective bloom development to occur these lower tidal ranges should have coincided with higher light levels. In actual fact the two factors are not independent because as noted above the *in situ* light field will also in part be determined by the tidal energy (Tett, 1990).

In Figure 9 the tidal height, irradiance and Dockhead chlorophyll *a* data are displayed on one graph. The pattern of bloom development was characterised by a broad spring bloom in late May and early June, followed by small blooms in June and then higher peaks in July and August. During spring and summer 1999 the tidal range in Southampton Water was below 2 m on five occasions as seen in Figure 2: the first week in April; second week in May; and during the second neap tide to occur in July, August and September. In the first week of April, although tidal energy was low, averaged daily irradiance was well below the threshold value and a bloom did not occur. The main spring bloom appears to have started following the low neap tide in the first week of May. At this stage average irradiance had been above 193 W m^{-2} day^{-1} for about a week. The sustained bloom corresponds to irradiance values above the threshold but also to a period when irradiance and weather patterns were variable. The bloom continued until it appears to have been finally dispersed by the high spring tide in the second week of June. This also coincided with a drop in irradiance. The irradiance data indicates a period of calm bright weather coinciding with the neap tides in June and early July. Peaks in chlorophyll *a* are present following these neap tides but the blooms are small, <5 mg m^{-3} chlorophyll *a*. While irradiance was high at these times the tidal range during these neap periods was above 2 m, suggesting that tidal energy probably limited the development of these blooms. Towards the end of July the neap tidal range was again less than 2 m. This also coincided with a period of good weather when the daily irradiance exceeded 300 W m^{-2} day^{-1}. The chlorophyll *a* concentration recorded at Dockhead reached over 8 mg m^{-3}. After this period the Ferry-Box data clearly show that each of the following three neap tides are associated with an increase in concentrations of chlorophyll *a* (Fig. 7). However the intensity of these peaks does not correspond directly to the tidal energy as indicated by the tidal height. The tidal height data suggests that the bloom at the end of August should have been the least dispersed and had the potential to become more intense than the preceding two blooms. The irradiance data however indicate that by this time of year the light levels were beginning to fall below the threshold value (Fig. 9).

These observations suggest that useful links can be made between the continuous Dockhead and Ferry-Box records and an interpretation based on simple threshold values of irradiance and tidal height. The threshold values give a useful indication of when blooms occur in the estuary. The extra information that is available in the Ferry-Box data relative to the Dockhead data is the spatial information which covers both the inner and outer estuary into the essentially open sea values recorded in the Solent. The wide range of chlorophyll *a* values seen in the estuary during blooms later in the year is seen in Figure 7. The maps shown in Figure 8 resolve the noise in Figure 7 into a consistent pattern reflecting changes both with time of day and position in the estuary.

In April (Fig. 8A) chlorophyll *a* concentrations are generally low (<2 mg m^{-3}) and patchy through the estuary with a gradient of increasing concentration towards the more open waters. This gradient may be due to a higher degree of productivity in these shallow waters, which contain less suspended sediment than the main channel of the estuary (Xiong, 2000). This chlorophyll *a* gradient is reflected in the daily variations seen in the Dockhead data. At the height of the spring bloom (Fig. 8B), concentrations of chlorophyll *a* are high throughout the estuary.

Later in the year the appearance of chlorophyll *a* becomes patchier. On 4 June 1999 concentrations of chlorophyll *a* are higher in the main channel of Southampton Water north of 50.8°N. In the higher salinity, more open waters, concentrations of chlorophyll *a* are below 1 mg m^{-3}. An extra factor in addition to tidal dispersion and light limitation may be important at this time of year. The pattern is consistent with the bloom being nutrient limited in high salinity water. The possibility of nutrient limitation during blooms associated with neap tides was indicated in measurements of nutrients and chlorophyll *a* made in the estuary on surveys conducted in 1998 (Xiong, 2000).

On 26 July (Fig. 8D) a further change in the pattern is visible. Like the bloom in June, the area of the bloom is confined to Southampton Water, but this bloom appears to show a marked change in intensity through the day as well as with position in the estuary. This change may be due to a shift in the composition of the phytoplankton population present in the estuary from one dominated by non-motile diatoms in May and June to one which has a large component of motile organisms (such as *Mesodinium rubrum*; Crawford, 1993). *Mesodinium rubrum* change their depth in the water column during the day in response to changing light conditions and to minimise their dispersion (Crawford & Purdie, 1992; Lauria et al., 1999).

Conclusions

The combination of the fixed-point sensor and the Ferry-Box system, in Southampton Water, made possible a resolution of shifts in bulk phytoplankton levels throughout the spring and summer 1999. The Ferry-Box data enabled the identification of the movement of the blooms within the estuary. The development of a bloom requires a threshold value of irradiance be exceeded. Subsequent development of a bloom then depends on a balance between irradiance driving plankton growth and tidal energy tending to disperse the bloom.

The observation of a main spring bloom, which developed against the high dispersive pressure of a spring tide, shows that a number of factors must be closely in balance in Southampton Water. For this bloom to occur both the actual growth rate of the phytoplankton present and the available *in situ* irradiance must have been high enough to produce a growth rate that was higher than the rate of dispersion. A well-known physical phenomenon in Southampton Water is the so-called double high water (Brown et al., 1989). There is no evidence at the moment that it has significant influence on the development of phytoplankton blooms. An idea that deserves further investigation is the possibility that the prolonged period of slack water makes Southampton Water more productive than other estuaries which have similarly large tidal ranges.

Overall the continuous data set presented here provides a context for more detailed process studies in the future. More experimental work is needed in the area to examine the light field in different conditions. The results should be incorporated into models to examine irradiance, turbidity and phytoplankton dispersion. An example is the model developed by Shi (2000) to reproduce the timing and magnitude of blooms in Southampton Water. To provide further information it is planned that oxygen, nutrients and photosynthetically active radiation (PAR) sensors be incorporated into the sensor arrays.

It is recognised that the chlorophyll *a* calibrations used in this work were not optimal. It is not clear at the moment to what extent the differences in concentrations estimated by the two systems resulted from simply the distance between the two systems, or applying inappropriate calibrations, or more subtle effects such as different degrees of fluorescence quenching. This remains to be investigated and in future work all of the fluorometers used will be inter-compared and calibrated using extracted chlorophyll *a* samples. A further technical improvement will be to tap the outflow of the Ferry-Box fluorometer so that chlorophyll *a* samples can be collected to provide *in vivo* chlorophyll *a* calibrations.

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We would like to thank the Red Funnel ferry group for allowing the Ferry-Box sensors onboard, and the crew of the Red Falcon for help during service and data collection visits. Also WS Ocean Systems Ltd for their help in designing, installing and maintaining both the Dockhead and Ferry-Box systems. The work was part funded in 1999 by the UK Department of the Environment Transport and the Regions (Contract CWO 805), the UK Environment Agency (Southern Region) and the UK Natural Environment Research Council. Particular thanks are due to H. Paterson and E. Ali for assistance with the calibrations, and to S. Alderson and M. Hartman for work on the plotting routines and scripts that they produced to handle such large quantities of data. Thanks also to R. Sanders and two anonymous reviewers for their thoughtful comments on the text.

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Identifying 'hot spots' of biological and anthropogenic activity in two Irish estuaries using means and frequencies

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Key words: hot spots, blooms, estuary, odds, point source

Abstract

The available data for two important Irish estuaries, Cork and Wexford harbours, were analysed to identify 'hot spots': locations where water quality variables are likely to differ from background levels. The approach taken reflects the limitations imposed by restricted spatial and temporal replication in the available datasets. Information for many estuaries may exist in such fragmented datasets. Averages drawn from small sample sizes are susceptible to extreme values. To lessen this problem, a novel approach was used: identifying locations where high measurements of a variable were relatively more frequent. The locations of relatively high chlorophyll measurements in Cork and Wexford harbours indicated estuarine origins for the majority of algal blooms. Nutrient cycling in Wexford Harbour appeared to be coupled with phytoplankton growth. The estuary acted as a source for dissolved inorganic nutrients during periods with low chlorophyll levels and as a sink during plankton blooms. High chlorophyll levels in Cork Harbour were generally associated with sub-surface samples in stratified water. Sources of ammonia and phosphate in Cork harbour appeared to result from direct anthropogenic input. Residual variation in biological oxygen demand reflected point pollution sources in both Cork and Wexford Harbours. Algal blooms were common in both estuaries, with 20% of chlorophyll *a* measurements exceeding 20 μg l^{-1} in each system. However, despite the presence of blooms and influences of point sources, there is currently little evidence for environmental impacts such as extensive deoxygenation. This conclusion is tentative, given the fragmented nature of the datasets. The locations of hot spots can be used to inform future research on potential impacts and estuarine function.

Introduction

Financial and logistic constraints partially restrict the number of estuarine systems that have been intensively studied. The complexity of estuarine processes revealed by concentrated research indicates that estuarine processes will vary temporally and from one system to another (Rendell et al., 1997). The application of ecological generalisations to systems that have not been studied in depth is a difficult process and there is still some requirement for major sampling programmes in individual estuaries (Nedwell et al., 1999). It is an oversimplification, however, to consider that no data exists for those systems that have not been studied in depth. Fragmented datasets, composed of small-scale research projects, statutory monitoring and individual surveys, are available for many estuaries. Such information is important on both a local scale, for characterising individual systems, and on a global scale as fragmented datasets are likely to be the only source of information about a large number of estuaries worldwide. This paper addresses some of the problems associated with using fragmented datasets to characterise two Irish estuaries, Cork and Wexford Harbours. An approach based on identifying locations where there was significant non-conservative behaviour of nutrients or plankton was used. Analyses of the frequency of high values were employed in addition to more traditional descriptions using comparisons of mean values. The estuaries studied here are good examples of systems where data exist without an overall context: some survey data are available,

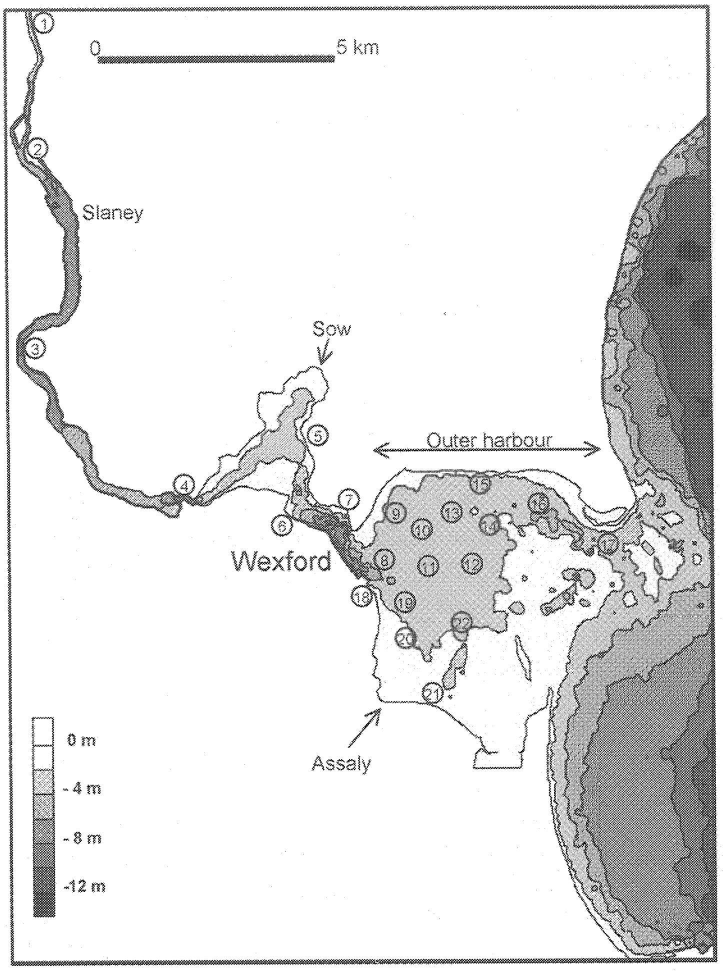

Figure 1. Sample site locations and bathymetry of Wexford Harbour. The River Slaney flows from the north west corner of the map.

but there are no summaries for Cork and Wexford Harbours in the published literature.

Conflicting demands are often placed on estuarine processes. For example, an estuary may simultaneously be the site of fisheries and waste disposal while having conservation value for migrating birds. As such, there is a need to characterise systems if real and potential anthropogenic impacts are to be judged in an appropriate context. One approach is to take a system variable and compare different sites or contrast different times. For example, the mean nutrient or chlorophyll concentration in separate estuaries can be used as a basis for comparison. This is an attractive approach, as the statistical theory relating to comparisons between means is widely understood (Jassby et al., 1997). A major drawback of comparisons of means is that concentrations of nutrients and chlorophyll in coastal waters frequently vary on daily and seasonal timescales (e.g. Tett & Wallis, 1978; Powell et al., 1989). This reduces the power of statistical tests, making it difficult to distinguish between separate means. For example, Johnson et al. (2000) concluded for a marine inlet that a sampling interval of less than a week would be needed to have confidence in detecting a difference of 50% between the mean chlorophyll concentration in different years. Jassby et al. (1997) gave further recommendations for spatial sampling intended to reduce the variation around estimates of the mean. Additional sampling issues are related to appropriate replication so that means are representative (Underwood, 1997) and that there is no psuedoreplication (Hurlbert, 1984). Fragmented datasets, by definition, lack the intensive sampling needed to have statistical power when comparing means. In addition, as estuaries are linked to their catchments by runoff, relatively high levels of nutrients and organic mat-

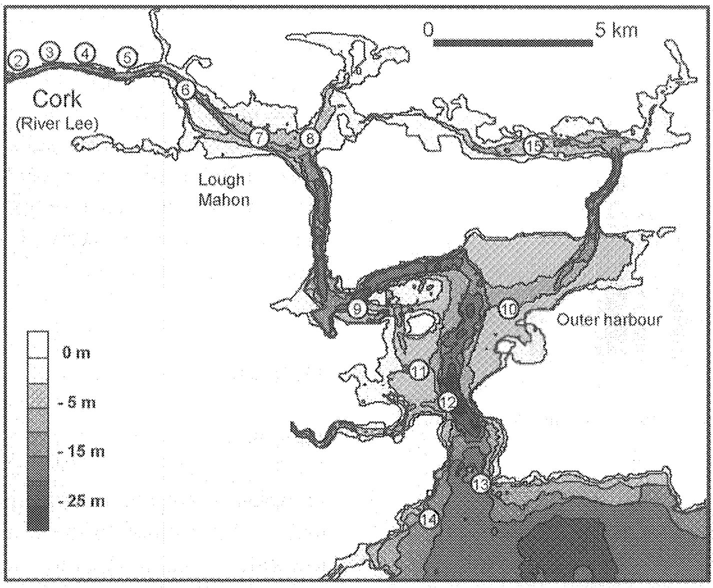

Figure 2. Bathymetry and sample site locations for Cork Harbour. The River Lee flows through Cork City in the north west corner of the map. The location of sample sites coded as site 1 varied. Most site 1 samples were taken from River Lee sites upstream of site 2, however a small number of samples were taken from smaller rivers in the catchment.

ter are to be expected in comparison to other coastal waters. Estuaries vary in the extent to which high nutrient concentrations are also associated with high chlorophyll levels (Cloern, 1999). Such considerations make it difficult to define reference levels that can be used for the identification of natural and impacted conditions.

In contrast to approaches based on estimations of mean concentrations for entire systems, it is generally easier to identify local (within system) phenomena that cause public health, amenity, economic or ecological problems (with problems in one sphere often having an impact in another area). Local phenomena in an estuary can be defined by the presence of non-conservative behaviour of water quality variables. Examples of relevant local phenomena are nutrient or plankton concentrations enhanced above the background level or oxygen deficient zones. These examples of non-conservative behaviour can be thought of as 'hot spots'. This paper argues that there are insufficient data to compare the means of water quality variables between estuaries or between years for the fragmented datasets of Cork and Wexford harbours. Hence, an approach is taken to identify the structure of hot spots within the available data. The locations of hot spots are used in an attempt to characterise Cork and Wexford harbours and to identify possible anthropogenic impacts on the two estuaries.

Study sites and available data

Wexford Harbour (52° 20.27′ N, 6° 22.24′ W) is in the South East of Ireland. The main flow into Wexford harbour is the River Slaney which widens to a shallow bay below Wexford town (Fig. 1). There are two smaller rivers, the Sow and Assaly, which enter the estuary at the north of the inner harbour just above Wexford and at the south of the outer harbour, respectively. The estuary is microtidal with a range of approximately 1.5 m on spring tides and 0.7 m on neaps. Figure 1 covers the extent of tidal influence on the Slaney with the catchment above sample station 1 being predominantly agricultural. There is a small amount of light industry and food processing associated with Wexford town. Effluent from the industrial sources and domestic sewage is discharged in the region of Wexford Town and in the south of the outer harbour. Habitats in the outer harbour are important for birds, particularly migrating waterfowl such as geese. The outer harbour is also an important site for mussel culture, with an annual production of approximately of 5400 tonnes.

Cork Harbour (51° 53.94′ N, 8° 27.68′ W) is a larger and more complex estuary than Wexford. Cork is the second largest city in Ireland and the estuary (Fig. 2) is the most industrialised in the country. There are industrial and domestic discharges at various points around Lough Mahon with further outfalls in the outer

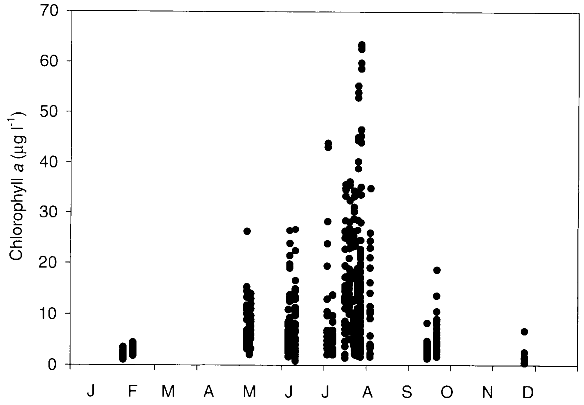

Figure 3. Chlorophyll *a* concentrations in samples from Cork Harbour, 1993–1999 (*n*=610).

estuary, the most important being near sample point 9 (Ringaskiddy). The main river entering Cork Harbour is the Lee, which flows through the centre of Cork City. The effects of other freshwater inflows on the estuary are thought to be relatively minor (Costello et al., 2000). The tidal range is greater than in Wexford, moving from about 2 m on neaps to 4 m on spring tides. Shellfish produced in the outer harbour are monitored and, under Department of Marine regulations, require purification in an approved plant for 48 h before sale for human consumption.

The data used in this study represent various surveys of each estuary carried out in the period 1993–1999. The earlier surveys were carried out by regional water laboratories, with results published by the Environmental Protection Agency. Later surveys, including some variables not measured previously, are from a programme sponsored by the Environmental Protection Agency. The datasets analysed are not exhaustive – they do not include a small number of pre-1993 surveys. However, the measurements represent coherent datasets where successful intercalibration has been carried out between participating laboratories (Costello et al., 2000). In general, there were between one and eight surveys carried out in separate estuaries each year. The datasets are based on 610 chlorophyll measurements in Cork Harbour and 514 measurements in Wexford Harbour. Sampling was concentrated in the summer months. Temporal patterns of chlorophyll concentrations suggest a seasonal pattern, but there was great variability in measurements within and between dates (Fig. 3). The datasets can be described as fragmented, as samples were not taken at regular intervals, with the overall effort varying between years. In addition, the exact number of sites visited and the number of water quality variables analysed may differ between surveys. As a consequence of the fragmentation, it is difficult to compare between years as samples may have been taken in different seasons. An estuary-wide average chlorophyll measurement for Cork or Wexford clearly has little meaning due to the bias towards summer months: a different average for the same period is likely if samples were stratified more evenly by season.

Methods

Field data

Samples were taken at the locations shown in Figures 1 and 2 close to the times of daytime high and low tide on each separate survey date. Surface water was collected in acid washed 2 l plastic bottles, kept in the dark, refrigerated (ca. 4 °C) and analysed within 24 h of collection. Salinity and temperature were measured with a field meter, calibrated against standard seawater from IOS in Godalming, Surrey, U.K. Where significant salinity or temperature stratification was detected, additional sub-surface samples were also taken (generally at depths of 2 or 4 m). Oxygen saturation (DO,%) was measured in situ with field meters calibrated for salinities from 0 to 40. Laboratory analysis of water quality variables used the following methods. Total nitrogen (TN) was converted to nitrate by a persulphate digest (Grasshoff et al., 1983) followed by automated cadmium column reduction to nitrite and colourmetric analysis in a Tecator flow injection analyser. Total oxidised nitrogen (TO×N, nitrate plus nitrite) was also analysed by flow injection analysis following cadmium column reduction, but without any digestion step (Grasshoff et al., 1983). Total ammonia nitrogen (TAN) was determined colourmetrically following oxidation with hypochlorite in an alkaline medium (Mackereth et al., 1978; Parsons et al., 1984). An ammonium persulphate–sulphuric acid digest in an autoclave (APHA 1985) was used to oxidise samples for total phosphorus (TP). Digested samples were brought to neutral pH with sodium hydroxide before analysis by the Murphy and Riley colourmetic method (Parsons et al., *op. cit*). Soluble reactive phosphorus (SRP) was determined with the Murphy and Riley colourmetic method on undigested samples (Parsons et al., *op. cit*). Silicate (Si) was measured using the oxalic acid-molybdate method (Parsons et al., *op. cit*). Phytoplankton biomass was estimated as chlorophyll *a* by spectrophotometer readings at 555 nm following cold methanol extraction and filtration (HMSO, 1980). Suspended solids (SS) were measured by collecting material on GF/C filters which had been pre-washed and dried (APHA, 1985). The biological oxygen demand (BOD) of sampled water was estimated from the change in oxygen concentration following dark incubation of subsamples at 20 °C for 5 days (APHA, 1985).

Statistical analyses

The central position of a distribution of sample values is commonly expressed using the mean. Where samples have particularly skewed distributions, for example chlorophyll *a* measurements, it is common to transform data to produce a more representative summary of the central tendency (often a ln $(1+x)$ transform in the case of chlorophyll). If a dataset is subdivided by location, a graph of means with relevant confidence intervals can provide a summary of the underlying spatial structure. The mean may not always be the most relevant statistic for water quality variables (Jassby et al., 1997; Moss, 1998). Extreme values of water quality variables, reflecting processes such as algal blooms, may be associated with environmental impacts and yet not reflected in mean values. Conversely, a problem associated with fragmented datasets is that, where sample numbers are small, the mean and confidence intervals will be strongly affected by any outliers. An alternative approach to summarising sample distributions is to look at the frequency with which a predefined threshold is exceeded. The effects associated with the magnitude of outliers are removed by looking at frequencies, but information on relatively high measurements is retained. The threshold value can be set at some level considered to represent an impact. An example threshold is the environmental quality standard of 5 mg l^{-1} for dissolved oxygen adopted as a guide for the passage of migratory salmonid fishes (Elliott, 1996). Of course, dividing the number of measurements exceeding a threshold by the total number of measurements provides an estimate of the probability that any single measurement will exceed the threshold.

Expressing the structure in water quality variable distributions using probabilities has an intuitive appeal. The odds of an event are calculated by dividing the probability of an event occurring (p) by the probability that the event does not occur ($1 - p$). Hence odds exceeding one have the simple interpretation that an event is more likely to occur than not. Odds statistics are commonly used in the analysis of frequencies (Sokal & Rohlf, 1995). The statistical significance of a single odds value can be assessed given an appropriate null hypothesis using binomial theory. It is possible to test for significant differences between two odds estimates by taking the logarithm of the ratio of the two estimates (the 'log–odds ratio') and testing for significance using the property that log–odds ratios are approximately normally distributed with mean of zero (Sokal & Rohlf, 1995).

Where locations with non-conservative behaviour of water quality variables were noted, principal component analysis (PCA) was used to summarise the patterns of abiotic data. Rank correlations were used to identify associations between the principal component axes and chlorophyll concentrations (Bakus, 1990; Muylaert & Raine, 1999).

Results

Wexford Harbour

The frequencies of chlorophyll *a* measurements above the median for the dataset and the values of mean chlorophyll *a* have similar spatial patterns (Fig. 4). Average values and odds of above median measurements were relatively low at the head of the estuary (site 1) and close to the mouth of the estuary (sites 15 and 17). Higher mean chlorophyll levels occurred at locations in the south of the outer harbour (sites 20 and 21) and in the Slaney above Wexford (sites 2 and 3). The same pattern was evident with odds ratios. Confidence intervals for the means from different sample sites are fairly broad, making it difficult to attach statistical significance to the observed patterns. In contrast, binomial tests of the odds at different sites show five significant departures from the expectation that the number of measurements above the median will equal the number of measurements below the median.

The relationships between abiotic variables at a site where high chlorophyll measurements were more frequent (site 3) are revealed by principal components analysis (Fig. 5). Components for TAN and SRP are adjacent, indicating that these inorganic nutrients tend to show similar distribution patterns in different surveys. In contrast, temperature and dissolved oxygen components are almost opposite those of TAN and SRP. Chlorophyll *a* concentrations were strongly correlated with the first axis of the PCA (r_S=0.6,

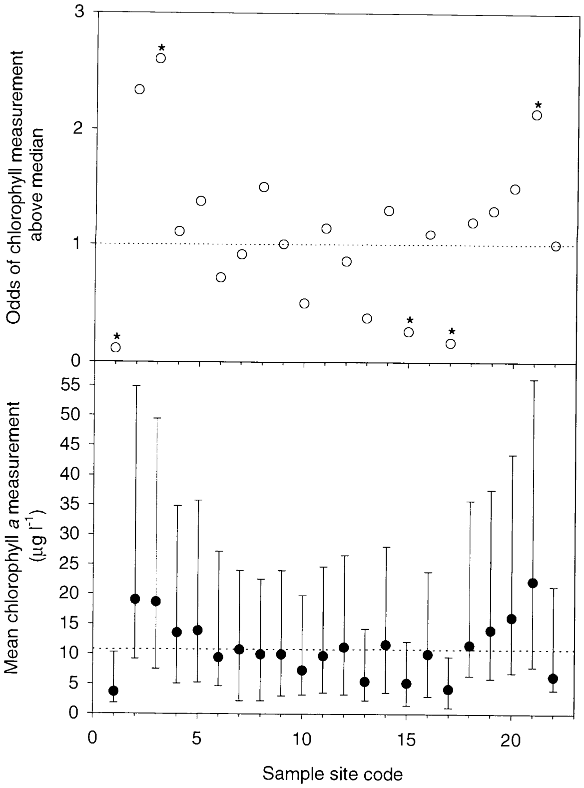

Figure 4. Variation between sample sites in the odds of a chlorophyll measurement above the median and mean chlorophyll measurements. The odds calculation is based on the median for all samples collected in Wexford Harbour (9.95 μg chl l^{-1}). Asterisks are used to identify odds that significantly differ from the expectation that half the measurements at a location will be above the median (binomial test, $p<0.05$). Chlorophyll distributions were positively skewed, so means were calculated on ln $(1+x)$ transformed data. Because means are shown on a linear scale after back-transformation the 95% confidence limits shown are asymmetric (the mean for the entire estuary is shown as a dotted line).

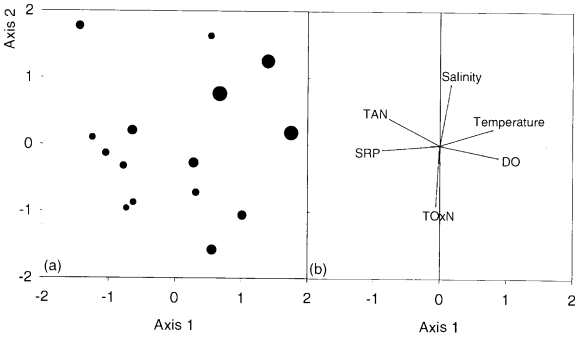

Figure 5. Principal component analysis of abiotic variables in samples from site 3 in Wexford Harbour. (a) Position of samples on the first two axis. Dot sizes represent the relative magnitudes of chlorophyll concentrations in samples. (b) Component plot for the first two axes. Axis 1 was associated with 45.6% of the variation between samples, while axis 2 explained 31.3% of the variation in the abiotic data.

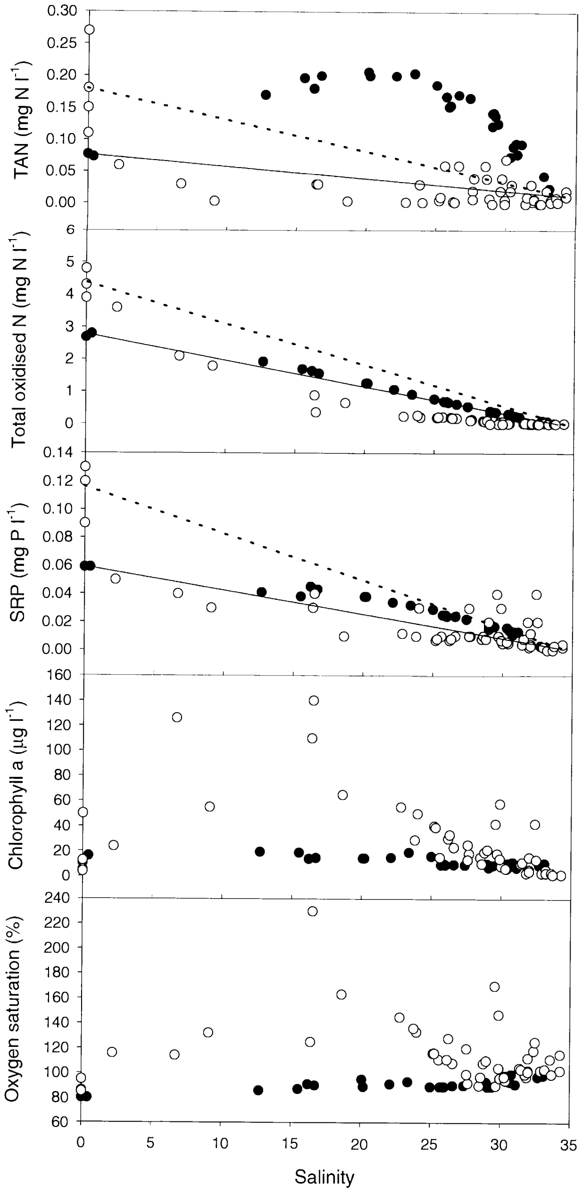

Figure 6. Salinity mixing diagrams for samples taken on 3 September 1998 (open circles) and on 10 September 1998 (closed circles) from Wexford Harbour. Guidelines for conservative nutrient behaviour are given between freshwater and marine end points with a dotted line for 3 September and a solid line for 10 September.

$p<0.05$) but only weakly associated with the second axis ($r_S=0.26, p>0.05$). This implies that blooms were associated with warmer temperatures, higher oxygen levels and lower concentrations of TAN and SRP. Components strongly associated with the second axis reflected a weak positive association between chlorophyll *a* and salinity ($r_S=0.36$, $p>0.05$) and a weak negative relationship between chlorophyll *a* and total

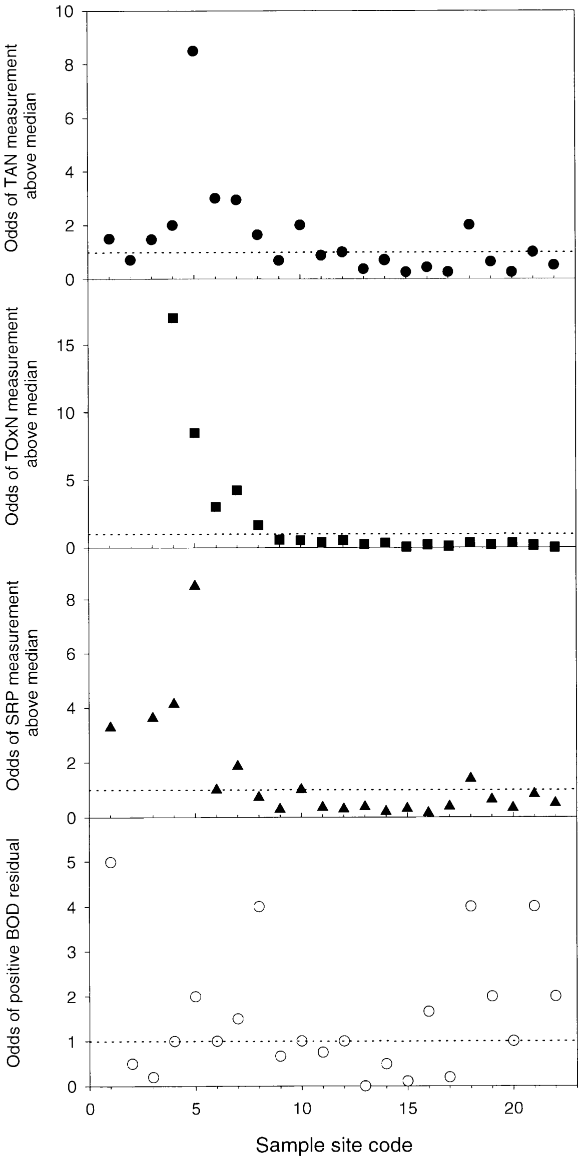

Figure 7. Odds of nutrient measurements above the Wexford estuary median and BOD residuals above the predicted BOD-chlorophyll relationship. Missing data points indicate that no odds could be calculated as all measurements were above the median.

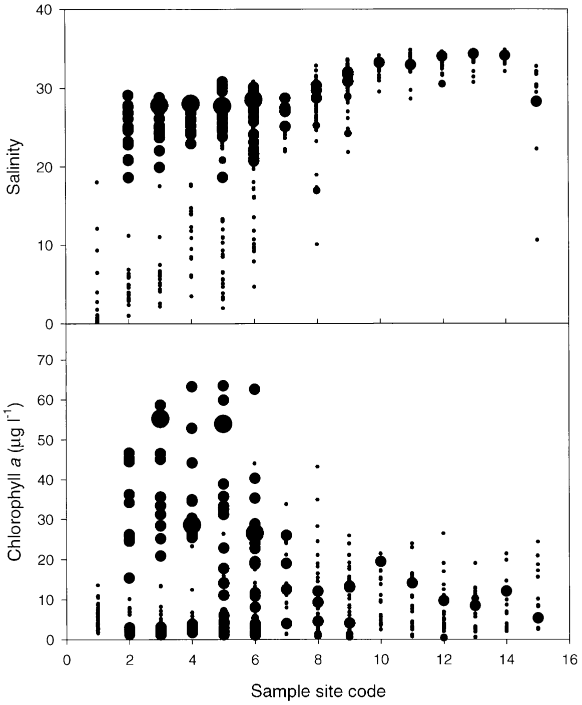

Figure 8. Variation in salinity and chlorophyll *a* with sample site in all samples taken from Cork Harbour. Data point size increases with sample depth.

oxidised nitrogen ($r_S = -0.37$, $p > 0.05$). Principal component analysis of the data from site 21 implied a similar structure to the relationships between abiotic variables. The strongest relationships between chlorophyll *a* and abiotic variables were negative correlations with TAN and SRP and a positive relationship with dissolved oxygen.

Salinity-mixing diagrams reflect the PCA results from individual sites (Fig. 6). Uptake of TAN, TO×N and SRP in the estuary was associated with relatively high oxygen saturations and chlorophyll *a* concentrations. Conversely, when there were relatively low chlorophyll *a* concentrations in all samples, oxygen levels were generally below 100% saturation and there was release of TAN and SRP within the estuary.

Summary plots for the odds of inorganic nutrient measurements above the estuary median reflect, on average, conservative dilution of TO×N from the peak values in freshwater entering the estuary (Fig. 7). In contrast, the odds of a relatively high measurement of TAN or SRP peak inside the estuary at site 5, with the probability of a high measurement also exceeding one at site 18. BOD levels are positively correlated with the chlorophyll *a* concentration in a sample. However, it is possible to correct for ambient phytoplankton concentrations by fitting a regression with chlorophyll *a* as the independent variable and BOD as the dependent variable. Positive residuals around the regression line are independent of the background level of chlorophyll and they therefore represent the influences of additional sources of organic carbon. Summary data for the odds of a BOD residual being positive indicate

Table 1. Spatial variation in mean chlorophyll *a* concentrations and the odds of a measurement exceeding the median value for Cork Harbour. Where fewer than five sub-surface samples were taken, records from a site were pooled across depths. Mean values were calculated using ln $(1+x)$ transformed data as the distribution of chlorophyll values was skewed

Sample site	Odds of chlorophyll *a* measurement above median		Mean chlorophyll *a*	
	Surface samples	Sub-surface samples	Surface samples	Sub-surface samples
1	0.38	–	5.38	–
2	0.60	1.67	4.79	13.04
3	0.45	1.67	5.01	14.36
4	0.60	1.67	5.89	15.44
5	0.33	1.50	5.25	11.47
6	1.20	1.30	7.61	10.32
7	1.86	–	10.21	–
8	1.73	–	8.80	–
9	1.56	–	8.31	–
10	1.09	–	8.58	–
11	1.07	–	7.73	–
12	0.94	–	6.34	–
13	0.92	–	6.08	–
14	1.20	–	7.30	–
15	2.25	–	10.33	–

Table 2. Odds that a BOD measurement from Cork Harbour will be greater than predicted on the basis of the chlorophyll *a* concentration of a sample. Odds from sites 11 to 15 were all zero

Sample site	1	2	3	4	5	6	7	8	9	10
Surface	0.05	1.80	3.00	1.80	2.40	3.00	0.21	0.42	0.27	0.00
Sub-surface	–	1.17	1.00	0.75	0.67	0.40	–	–	–	–

additional sources of organic carbon near sites 1, 8, 18 and 21.

Cork Harbour

In contrast to Wexford Harbour, there is evidence for strong and persistent stratification at sample sites in the upper estuary near Cork City (Fig. 8). Sub-surface samples from sites 2, 3, 4, 5 and 6 were always more saline than surface samples taken at the same time. The average increase in salinity associated with sub-surface sampling was 15.17 (S.E. 0.82). Stratification appeared to be associated with high chlorophyll *a* concentrations, with the odds of an above median measurement exceeding 1 in sub-surface samples from sites 2, 3, 4, 5 and 6 (Table 1, median chlorophyll concentration=5.83 μg l^{-1}). Pooling the data from sites 2 to 6 by sample depth shows that relatively high chlorophyll *a* measurements were significantly more likely in sub-surface samples than in surface samples (log–odds ratio test, z=3.20, p<0.01). Relatively high chlorophyll measurements were also more frequent at site 15, although peak chlorophyll values only occurred at sites 2–6 (Fig. 8). As with the samples from Wexford, it is possible to correct BOD measurements for ambient chlorophyll. The odds that a residual BOD measurement is positive after chlorophyll correction vary with sample location and depth (Table 2). Relatively high corrected BOD measurements are three times more likely in surface samples from sites 2 to 6 in comparison to sub-surface samples from the same locations (log–odds ratio test, z=3.21, p<0.01).

The link between sample depth and salinity is clear from the positioning of the relevant components in a PCA plot (Fig. 9). The clustering of components for nutrients suggests that surface waters have relatively higher concentrations. Chlorophyll *a* measurements have a strong negative correlation with the first axis, reflecting the odds ratio statistics ($r_S = -0.71$, $p < 0.001$). There was a slightly weaker, but still significant relationship between estimated phytoplankton biomass and the second axis of the PCA ($r_S = 0.45$, $p < 0.001$). This may reflect some seasonal variation in the estuary. With sample day converted into minimum number of days until January 1, counting forwards or backwards, there was a significant positive relationship between chlorophyll *a* and sample day ($r_S = 0.52$, $p < 0.001$, see Fig. 1).

Salinity-mixing diagrams reflect the divergent behaviour of SRP and TAN when compared to TO×N and silicate in the PCA plots (Fig. 10). In contrast to the near-conservative behaviour of TO×N and silicate, there were increases of both TAN and SRP at salinities around 10. The presence of peak TAN and SRP concentrations at intermediate salinities suggests sources for these nutrients within the estuary. The inorganic nutrients at sites 2–6 tended to have inverted profiles, with higher concentrations in surface waters overlying lower concentrations at depth.

Discussion

Wexford Harbour

It was possible to identify hot spots; locations associated with elevated concentrations of water quality variables beyond the background level expected from simple dilution processes. Phytoplankton blooms were more likely to originate within the estuary than to enter the estuary from marine or freshwater boundaries. Exceptional algal blooms could occur, with chlorophyll *a* concentrations occasionally exceeding 100 μg l^{-1}. These blooms depleted inorganic nutrients and generally increased dissolved oxygen concentrations. Nutrient cycling at intermediate salinities was generally coupled with phytoplankton blooms; uptake during blooms contrasted with release of TAN and SRP when chlorophyll concentrations were lower. It seems likely that blooms would be stimulated by increased water residence times in Wexford Harbour. The positive association between salinity and chlorophyll suggests that lower freshwater flows in the Slaney may lead to blooms (although the relationship is weak). A stronger indication of the role of freshwater input is given by a negative relationship between the available data for daily gauged flow at a site further up the Slaney and the mean chlorophyll concentration at sites 2 and 3 (Pearson correlation coefficient $= -0.81$, $n = 6$, $p = 0.05$). The data are, unfortunately, insufficient to separate the effects of seasonal changes in flow from day to day changes associated with variations in rainfall. Neap tides will also reduce flushing in the estuary. In this respect, Figure 6 may reflect the influence of variations in tidal flushing. Surveys made just after neap tides (September 3) show evidence of nutrient depletion and plankton blooms. In contrast, the chlorophyll peak is not present and there is release of nutrients following a spring tide (September 10). Neap tide increases in chlorophyll have been reported for other estuaries, generally associated with increases in stratification (Webb & D'Elia, 1980; Cloern, 1984). There is little evidence for stratification on neap tides in Wexford Harbour but more data are needed to make definite conclusions about the effects of spring-neap tidal cycles.

Sites where relatively high TAN and SRP measurements were more frequent cluster around Wexford Town and the south side of the outer harbour. The frequencies of positive BOD residuals show a similar pattern with the exception of site 1, where 83% of chlorophyll corrected BOD values were positive. These anomalies appear to reflect the influences of point sources of sewage in the estuary. Surveys of faecal coliforms indicate that site 1 has the highest counts in the estuary (median 5550 cells $\times 10^{-2}$ ml^{-1}, Neill, 1995). Sample sites near Wexford town and the south Harbour also appear to have coliform counts elevated above background levels.

Despite the presence of hot spots for plankton blooms and anthropogenic pollution in Wexford Harbour, these phenomena are not, as yet, linked to serious ecological or economic problems. Degradation of blooms or BOD loads did not seem to cause serious oxygen depletion. Measured oxygen concentrations did not fall below 40% saturation with a mean saturation of 101.22% ($n = 670$, S.D.=15.97). This assessment of oxygen levels is, however, conditional on the fact that most samples were taken during the day whereas oxygen depletion is more likely at night. It is of interest that nutrients can become depleted close to detection limits during a bloom. If this reflects nutrient limitation of blooms, then changes in nutrient export from the catchment or Wexford Town are likely to affect the magnitude of blooms.

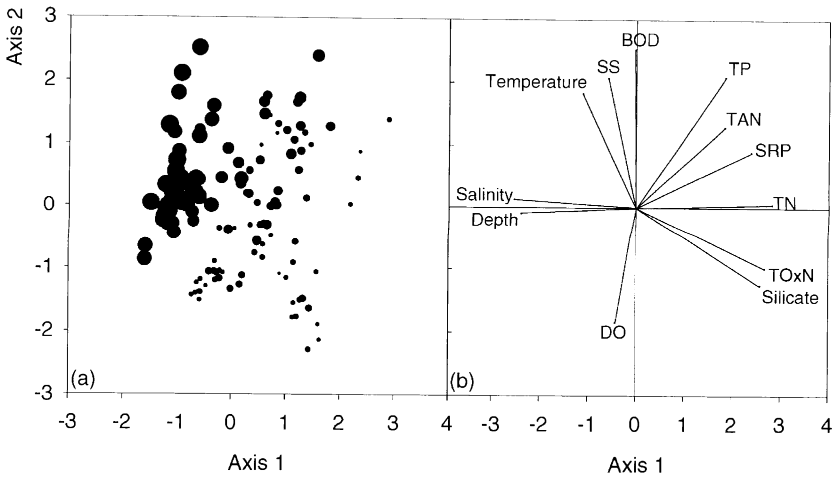

Figure 9. Principal component analyses of abiotic data from samples taken at sites 2–6 in Cork Harbour. (a) Sample positions on the first two axes of the PCA. Dot size increases with chlorophyll concentration. (b) Contribution of the abiotic variables to the first two axes of the PCA. Principal component 1 explained 44.8% of the variation between samples, with the second axis associated with 25.0% of the variation.

Cork Harbour

As in Wexford Harbour, high chlorophyll concentrations were more likely in certain locations within Cork Harbour and there was evidence for point sources of organic material and nutrients. Analyses using both odds ratios and means suggest that the most common algal blooms occur in the sub-surface saline water between Cork City and Lough Mahon. The sub-surface chlorophyll maxima could be the result of in situ growth or accumulation of phytoplankton cells. It seems likely that in situ photosynthesis contributes to the high sub-surface algal concentrations, as there is a positive correlation between dissolved oxygen levels and chlorophyll (r_S=0.30, $p<0.001$). Hence the relatively high chlorophyll levels are not solely the result of sinking surface plankton or advective accumulation from elsewhere in the estuary. A number of mechanisms could be involved in the net phytoplankton growth in the sub-surface layer. Simple growth rate responses to optimum light and nutrient conditions seem unlikely as the surface layer had both higher light levels and nutrient concentrations. There may have been photoinhibition at the surface, but this seems unlikely given the high turbidity in the estuary: mean secchi disk depth at salinities below 30 was 1.52 m (n=253, S.E.=0.0027). The estuarine circulation implied by salinity stratification in Lough Mahon may have caused differences in residence time with depth: a rapidly flushed brackish surface layer over more slowly mixed sub-surface waters. Increased residence time in the sub-surface layers would allow more time for in situ photosynthesis and algal growth. Other alternative explanations include osmotic stress of surface plankton (Muylaert & Raine, 1999) or vertical differences in grazing pressure. Clearly further data are needed to resolve the factors influencing sub-surface chlorophyll peaks.

Salinity-mixing diagrams show concentrations of TAN and SRP in the surface waters at sites close to Cork City rising above the levels in freshwater inflows. The shape of the mixing curves and the inverted nutrient profiles suggest that the non-conservative behaviour of SRP and TAN is linked to low salinity point sources of municipal and industrial waste between Cork City and Lough Mahon. This conclusion is supported by the relatively high frequency of positive residual BOD measurements in the surface waters of sites 2–6. There is also evidence for more frequent above median chlorophyll measurements at site 15, although maximum values at this site not reach the levels recorded in sites 2–6 (Fig. 8). The above median chlorophyll values may reflect increased residence times in the partially enclosed water body around site 15. Site 15 is some distance from major population or industrial centres and there was no evidence for additional BOD loads.

The stratification and point sources complicate the interactions between phytoplankton and nutrients and hence Cork Harbour does not show the same sort of nutrient–plankton coupling as Wexford. Relatively high chlorophyll concentrations were, however, common in both estuaries, with 20% of chlorophyll *a* readings in each system exceeding $20\ \mu g\ l^{-1}$. Low silicate

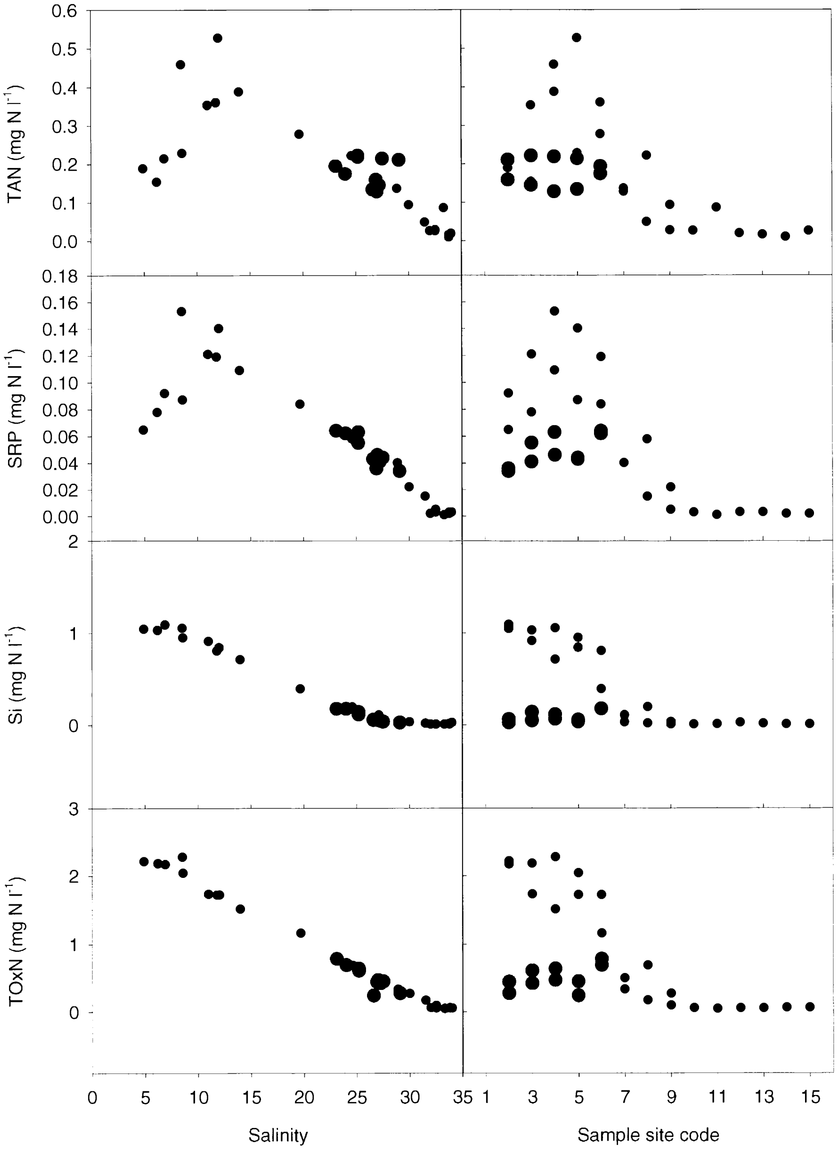

Figure 10. Salinity and location plots for dissolved organic nutrients on 20 July 1998 in Cork Harbour. Larger dots represent sub-surface samples.

availability relative to inorganic nitrogen (Fig. 10) may potentially favour growth of flagellates instead of diatoms (Egge & Aksnes, 1992). Given the additional BOD loads and presence of high chlorophyll levels in Cork Harbour, there may also be ecological problems associated with deoxygenation. There was some evidence for occasionally low oxygen levels, with seven readings below 50% saturation. However, the mean oxygen saturation was 88.01% (*n*=556, S.D.=14.31). As in Wexford Harbour, measured concentrations do not reflect any night time oxygen minima as sampling was generally during the day. Point pollution sources affected the surface waters of the River Lee and Lough Mahon. Discharge to surface waters may mitigate potential ecological impacts as stratification leads to the additional nutrient and BOD loads being separated from the locations of chlorophyll peaks in the sub-surface layers.

Problems with fragmented datasets

Missing data will always hinder analyses of fragmented datasets. As an example, temperature is associated with blooms in both estuaries, but it is not currently possible to separate the seasonal signal from day to day variability associated with fine weather or low freshwater flows. In some senses, one can never have sufficient data and there is always a possibility that a different picture would emerge from a different or more intensive sampling programme. Records from automated samplers tend to emphasise the importance of high-resolution sampling to ecosystem studies (Taylor & Howes, 1994). Two kinds of bias can be distinguished with respect to fragmented datasets. Sampling bias occurs when samples are absent from certain areas or times. Structural bias refers to cases where samples are not evenly dispersed in space and time within the fragmented dataset.

Little can be done to counter sampling bias except to give information about sampling frequencies and locations (Figs 1–3). Care must be taken not to extrapolate conclusions beyond the scope of the fragmented data set. Of course, these concerns apply to most empirical science. With respect to the datasets presented here, the presence of sampling bias is addressed by avoiding comparisons of years or estuaries, stressing the increased frequency of summer sampling and making conclusions about dissolved oxygen levels conditional on the time of sampling. In contrast, structural bias can be estimated and removed from datasets if necessary. An example of structural bias would be more frequent sampling in certain parts of an estuary during periods when chlorophyll concentrations were high. This could increase the apparent frequency of blooms in the selectively over-sampled sites. Examining the correlations between sample frequency at each site and the mean chlorophyll in separate surveys can check such an effect. Any over sampled sites will have a stronger positive correlation than the other sampling locations (under sampled sites will have a more negative correlation than other locations). The bias can be removed by randomly deleting data so that sampling frequencies during high chlorophyll events are balanced across all sites. This analysis was carried out for the Cork and Wexford datasets, but there was no evidence that more frequent sampling of hotspot sites during periods with elevated chlorophyll concentrations biased the identification of hotspots. The data were evenly balanced among sampling locations and survey dates within the fragmented datasets.

Conclusions

It was possible to partially characterise Cork and Wexford Harbours by looking for hot spots of non-conservative behaviour associated with algal blooms or anthropogenic inputs. Both estuaries contained areas where the likelihood of a plankton bloom was increased and proximity to urban areas was associated with detectable increases in nutrients (c.f. Puget Sound, Mackas & Harrison, 1997).

Use of odds to summarise trends has a simple appeal and interpretation. It is easy to communicate that if the odds of an event exceed 1, the event is more likely to occur than not. Another way of communicating this is by thinking of odds as a measure of risk. An odds ratio extends the concept to comparisons of the relative chance of an event in two different circumstances. Odds are a parsimonious way of summarising environmental data. For example, part of the rationale for reporting mean nutrient levels in a system is that higher means imply greater probabilities of algal blooms. However, mean concentrations do not themselves directly yield probabilities; it may sometimes be simpler to use the probabilities of events such as algal blooms directly.

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Distribution patterns of nutrients and symptoms of eutrophication in the Rio de la Plata River Estuary System

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Key words: nutrients, mixing diagrams, river flow, stratification, eutrophication assessment, Rio de la Plata estuary

Abstract

In this paper we discuss nutrient dynamics and the effects of eutrophication in the Rio de la Plata River Estuary System since 1980. The tidal river was characterized by high suspended particulate matter (SPM), nutrients, and N:P ratio (>25), moderate chlorophyll *a* values, an inverse relationship between SPM and phosphate, and cianobacteria blooms. Seaward of the salinity front, where both SPM and nutrient concentrations are lower and chlorophyll *a* values greater, the net ecosystem metabolism is positive. Permanent stratification controls nutrient, organic matter and oxygen dynamics leading to biological stress and hypoxia below the halocline. Non conservative behavior of nitrogen in the estuary leads to a low N:P ratio (<3) because of both phytoplankton assimilation of nitrogen and denitrification, and benthic flux of phosphate. Periodic nuisance and toxic blooms occur at high salinities along the Uruguayan coast (Canal Oriental). Over the recent decades, changes in freshwater inflow, point and non-point nutrient load, and stratification, triggered by ENSO events, seem to have controlled the ecosystem metabolism, nutrient-oxygen dynamics, and the development of harmful blooms. The assessment of the regional and overall ranking of eutrophic conditions determines that the system is moderately eutrophied. Nevertheless, increasing trends in quantity of freshwater and nutrient loads, and the low potential to dilute and flush nutrients, suggest that the Rio de la Plata, in particular the Canal Oriental is prone to worsening eutrophication conditions like oxygen stress and harmful blooms.

Introduction

Estuarine systems are exposed to cultural eutrophication which leads to an increase of productivity, hypoxia or harmful blooms (Justic' et al., 1995; Nixon, 1995; Rabalais et al., 1999; Rabalais & Turner, 2000). Two causes together determine the level of expression of eutrophic conditions, the natural susceptibility to developing problems caused by the natural flushing characteristics and nutrient inputs (Bricker et al., 1999). Primary symptoms of eutrophication i.e. high phytoplankton biomass, are often related to secondary symptoms like low oxygen contents. Pioneer articles of Webb & D'Elia (1980), Officer et al. (1984), and the review of Sinclair (1981), showed the importance of vertical stability in oxygen and nutrient distribution, benthic fluxes, exchange rates across the halocline, the temporal patterns and magnitude of phytoplankton biomass and production. Stratified systems are highly susceptible to oxygen stress below the halocline as a response to nutrient over-enrichment and the consequent increase of productivity and organic matter respiration. Rabalais et al. (*op.cit.*) have shown how high nitrogen input and saline stratification associated to the Mississippi river discharge, increased primary production rates and developed very extended hypoxic areas in the Gulf of Mexico.

The Paraná-Uruguay system water discharge is the world's sixth largest (Shiklomanov, 1998). Its economic value for transportation, fisheries, and tourism is enormous, and its sustainable development an im-

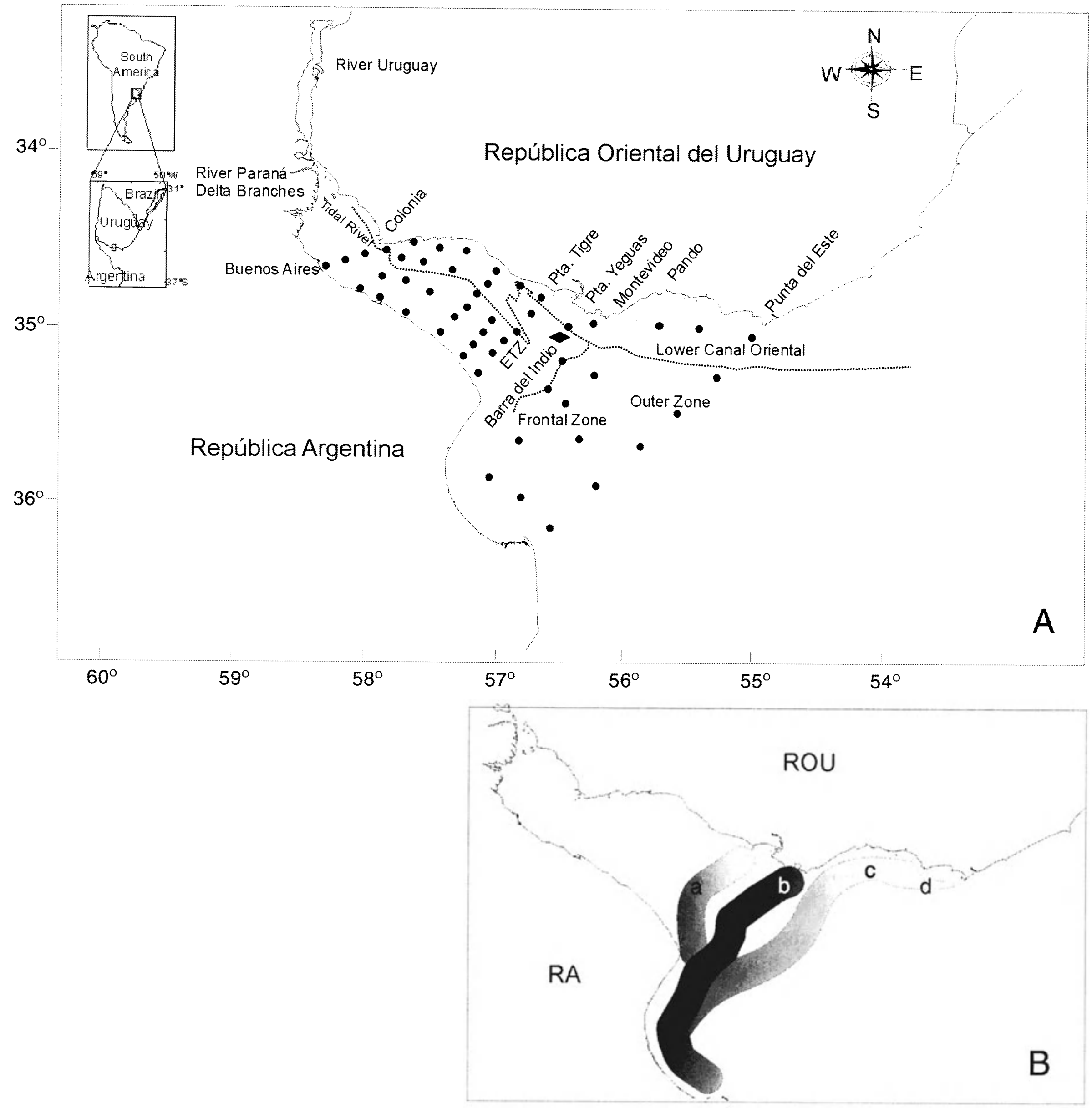

Figure 1. Rio de la Plata location (A), cited regions and geographical names (ETZ: Estuarine Turbidity Zone). Estuarine plume extension (B) along the Canal Oriental for different River Uruguay flows; (a) ~1500 m^3 s^{-1} (November 1999), (b) ~5500 m^3 s^{-1} (mean), (c) ~11 400 m^3 s^{-1}(August 1987), (d) 20 000 m^3 s^{-1} (strong El Niño events). Data from Nagy (1989); Nagy et al. (1997); Framiñán et al. (1999), and this work.

perative (Wells & Daborn, 1997). Because of its population, and its industrial and agricultural importance, its basin is very important in Latin America. Its fertile temperate grasslands ('Pampas'), that sustain about 20% of the world's cattle stock, have long been subjected to mounting human pressures, i.e. fertilization, induced erosion because of bad agricultural practices. In addition to that, the increase of El Niño-Southern Oscillation (ENSO) related variability and extreme events enhanced the vulnerability of soils to erosion and increased leaching.

Non-point sources of nutrients are the main input of nitrates, whereas point sources from Buenos Aires (10 *Million* inhabitants) and Montevideo (1.5 *Million* inhabitants) increase nitrogen and phosphorus. The latter reached, according to Pizarro & Orlando (1985), about 25% of the total nutrient load to the system in the early 1980s.

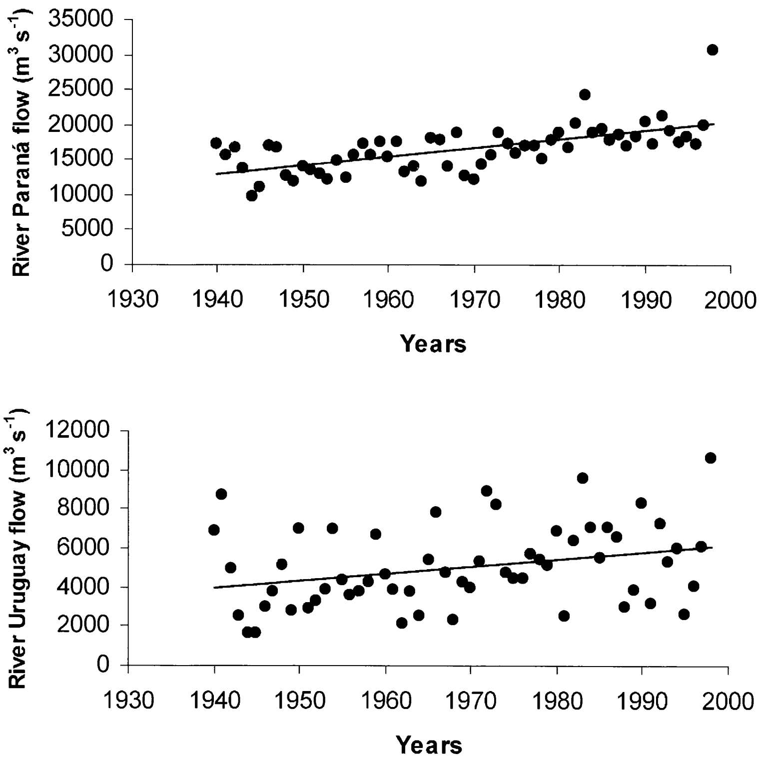

Figure 2. River flow of the Rivers Paraná and Uruguay since 1940. Both rivers show increasing trends and interannual variability, especially the latter, in recent decades. Sources: INAA, Argentina and SOHMA, Uruguay.

The geographic extent and water dynamics of the Rio de la Plata suggest that it is not yet in danger. However, the balance of evidence suggests that the health of the Rio de la Plata is declining due to both anthropic and natural causes (Gómez-Erache et al., 2001a). The latter, partly attributable to El Niño events, have reduced the yearly mean surface salinity off the Uruguayan coast (Nagy et al., 1996) and increased saline stratification. Also, extensive cyanobacteria and toxic blooms have occurred since 1980 (Méndez et al., 1997; Gómez-Erache & De León, 2000). This, together with the global tendency of coastal systems to become heterotrophic in the long-term (Smith & Hollibaugh, 1993), suggest that we must care for this system if we want to avoid an environmental crisis in a few decades.

Our goals are: (i) to analyze some nutrient source-sink processes in order to assess the regional and overall susceptibility to the eutrophication the Rio de la Plata has, and (ii) to review the main effects of eutrophication recently reported in the literature, in technical reports, as well as unpublished data.

Table 1. Study sites area, average depth, volume, and prevailing stratification (slightly stratified-B, and highly stratified-C). Values are averages and vary over all estuarine variability time-scales

Study site	Area $\times 10^3$ km^2	Depth m	Volume $\times 10^{11}$ m^3	Stratification
Tidal River	15	5	0.8	Freshwater
Frontal Zone	60	8	0.5	B and C
Canal Oriental	5	17	0.9	C and B
Outer Zone	10	14	1.4	B and C

Table 2. Prevailing wind directions, in percentage, at the Montevideo-Punta Carretas meteorological station for the period 1977–1986. Modified from Nagy et al. (1997)

Direction	Fall	Winter	Spring	Summer
N-NE	30	27	29	30
E-SE	26	27	41	44
S-SW	20	20	16	14
W-NW	23	25	13	12

Description of sites studied

General morphology, hydrology, and hydrography

The Rio de la Plata river estuary system (Lat. 35° 00′–36° 10′ S, Long. 55° 00–58° 10′ W) Southeast South America, covers an area of 36 × 10^3 km^2 (see Fig. 1). We divide the system into the tidal river and the estuary. The latter comprises three zones (as shown in Table 1): frontal zone, marine, and lower Canal Oriental or estuarine plume (see Fig. 1b). The rivers Paraná (75%) and Uruguay, the total drainage of which covers ~3.2 × 10^6 km^2 (the fourth in the world), feed the freshwater inflow to the system. A hydro-climatic shift occurred in the Rio de la Plata basin in the early seventies (García & Vargas, 1998), and both mean yearly discharge and fluctuations have increased (see Fig. 2), partly associated to ENSO related variability (Nagy et al., 1997). Seasonal and interannual freshwater discharge (Q_V) typically varies between 22 000 and 28 000 m^3 s^{-1} on both seasonal and interannual time-scales (mean Q_V ~26 000 m^3 s^{-1} for the nineties), with extreme values during El Niño (>30 000 m^3 s^{-1}) and La Niña (<20 000 m^3 s^{-1}). Climatologically, the Rio de la Plata has maximum discharge in March–June and September–October. The minimum discharge usually occurs around January. Northwesterly winds prevail in Fall–Winter and eastern and southeasterly winds prevail in Spring-Summer (as shown in Table 2). The tide is typically semidiurnal and has a maximum range of 0.4 m off the northern coast and 1.0 m off the southern one for the same transverse section because of Coriolis force (Balay, 1961; CARP, 1990). Tidal current speeds vary from 0.3 to 0.8 m s^{-1}. According to the terminology of the stratification-circulation (S-C) pattern classification of Hansen & Rattray (1966), the prevailing S-C type in the Rio de la Plata is 2b or partially mixed estuary with appreciable stratification (López Laborde & Nagy, 1999). This pattern varies with river flow, wind and tide-height: depth ratio from 1b (net flow is seaward at all depths) to 4 (salt wedge). Winds and river discharge show similar temporal patterns in relation to their effect on salinity fields distribution, with onshore winds and low discharge during Summer, and offshore winds and high discharge in Fall–Winter (Framiñán & Brown, 1996; Guerrero et al., 1997; Nagy et al., *op. cit.*). The mean water temperature (1981–1987) is ~15 °C (CARP, *op. cit.*)

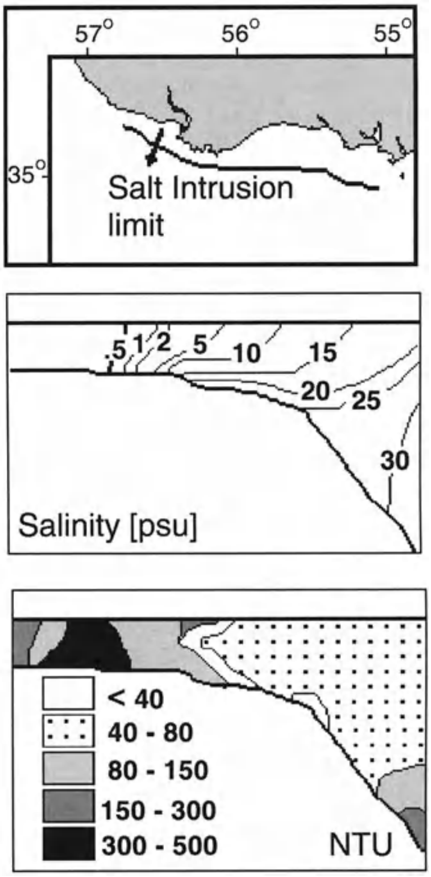

Figure 3. Typical location (measured in April 1986) of the salt intrusion limit and salinity fields (top and middle), and the turbidity maximum (in Nephelometric Turbidity Units) along the Canal Oriental. Modified from López Laborde & Nagy (1999).

Study sites

The tidal river (3–6 m depth) is a freshwater (<0.5 psu), turbid (photic layer <1 m), and river-dominated environment characterized by two main tidal channels separated by a network of sandy bars (Nagy et al., *op.cit.*; López Laborde & Nagy, *op.cit.*). An estuarine turbidity maximum (ETZ) connected with gravitational circulation and clay particles flocculation develops associated with the salt intrusion limit (López Laborde and Nagy, *op.cit.*; see Fig. 3); it's accumulation and migration is also related to the dynamics resulting from the asymmetry of the tidal wave, but with short tidal excursion (~3–5 km). The mean upstream limit of the saline intrusion (average 0.5 psu bottom isohaline) is located at the transverse section Punta Yeguas (for Q_V ~25 000 m^3 s^{-1}). The frontal zone (5–10 m depth; photic layer= 1–5 m) is characterized by the turbidity front (ETF) located on an average close to the 5 m isobath 'Barra del Indio' (Framiñán & Brown, *op.cit.*). The Canal Oriental is a deep depression (6–25 m depth) along the northern coast divided in a proximal (<10 m depth, at the estuarine front) and distal or lower portion (>10 m depth). The discharge of the River Uruguay explains most of the variability of salinity at Montevideo and the estuarine plume extension along the Canal Oriental (see Fig. 1b). The S-C pattern varies from 2b to 4 with the increase of River Uruguay flow. The outer or marine zone (10–20 m depth and ~150–200 km width) is a low turbid environment (photic layer= 5–12 m), highly stratified during flood periods and slightly stratified during low discharge, i.e. in Summer.

Materials and methods

Sampling

Because of the geographic extent of the Rio de la Plata, a large number of observations are needed to obtain a synoptic description of the whole system and to solve spatial and temporal variability. Data were taken during: (i) 15 cruises in the open waters of the tidal river and estuarine regions, between March 1981 and August 1987 (see Fig. 1; data from CARP, 1990); (ii) two cruises along the Uruguayan coast (Canal Oriental) in January and November 1999 (EcoPlata Project); (iii) one fixed point measurement carried out over 36 h at the estuarine turbidity maximum (ETZ) in November 1989. Long-term monthly data for river flow (Rivers Paraná and Uruguay) and coastal salinity at Montevideo were supplied by Instituto Nacional del Agua y Ambiente (INAA) from Argentina and Servicio Oceanográfico, Hidrográfico y Meteorológico de la Armada (SOHMA) from Uruguay.

Measurements

Water samples were collected for salinity, dissolved oxygen, pH, pigments, suspended matter and nutrients, with Van Dorn and Niskin bottles from the surface layer, above and beneath the halocline, and 1 m above the bottom. The number of samples varied from two to six according to depth and vertical salinity profile. Vertical profiles of salinity and temperature were determined at each meter with a YSI-33 salinometer. During the fixed station, samples were taken every hour for hydrographical and chemical variables.

Salinity was measured with a Beckman induction salinometer. Water transparency was measured with a 30-cm diameter Secchi disk. Samples for chlorophyll analyses were filtered using Whatman-GF/C filters and extracted with 90% acetone for 24 h in a cold, dark place. Chlorophyll concentration was determined spectrophotometrically with a Beckman UV-26 spectrophotometer, following UNESCO (1969) equations for samples before 1989, and Jeffrey & Humphrey (1975) equations for those taken in 1999. Nutrient concentrations were analyzed with Beckman UV-26 and DU-650 spectrophotometers, according to Strickland & Parsons (1972) for nitrate, nitrite, phosphate and silicate, and for ammonium according to Koroleff (1976). Dissolved oxygen and suspended particulate matter (SPM) were measured also according to Strickland & Parsons (*op. cit.*). Turbidity (Nephelometric Turbidity Units) was measured with an HACH-2100A turbidimeter.

Despite their complexity (Humborg, 1997), mixing diagrams were used to investigate the general longitudinal distribution patterns of dissolved nutrients within the area. They indicate whether an element is removed or added during the mixing of waters of different salinities. The necessary assumption is that freshwater and marine end-members are set constant in time (Liss, 1976) over the residence time of the water in the estuary. If this condition is not satisfied, non-linear relationships are expected (Cifuentes et al., 1990). Both residence time of water in the system and stoichiometric calculations of net ecosystem metabolism (NEM) and denitrification, were estimated following LOICZ procedures (Gordon et al., 1996). The former by using a simple steady-state one box two layers model and the latter for one layer. Preformed nutrient values were estimated by plotting the apparent oxygen utilization (AOU or anomaly) against measured nutrients.

Eutrophication susceptibility and conditions were assessed by using the methodology followed in the National Estuarine Eutrophication Assessment (Bricker et al., *op. cit.;* Ocean Studies Board, 2000). In order to give a large-scale paramount of the open waters, coastal eutrophied locations were not retained to calculate the eutrophication scores.

Results

Hydrology and hydrography

The mean annual river flows (1980–1998) were ~19 000 m^3s^{-1} (17 000 to 30 700 m^3s^{-1}) for the

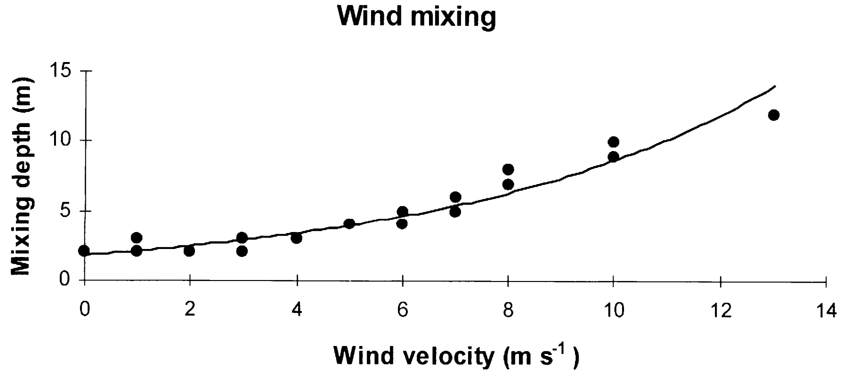

Figure 4. Relationship between average wind speed (12 h before) and the depth of the mixed layer (depth of the halocline) during two successive cruises in May and June 1981.

River Paraná (Q_P), and ~6000 m^3s^{-1} (2500–10 600 m^3s^{-1}) for the River Uruguay (Q_U). The latter showed greater seasonal and interannual variability and more rapid changes. Several El Niño events (1983, 1987, 1992, 1997) have induced greater seasonal and interannual variability and the increase of runoff (see Fig. 2). Strong La Niña events (1988, 1994, 1998) have partly counteracted the increase of river discharge, but also contributed to a greater variability over the involved timescales. Extreme seasonal inflow values have been in April–May 1998 (~60 000 m^3s^{-1}), May–July 1983 (~45 000 m^3s^{-1}), January 1998 (41 000 m^3s^{-1}); January 1986 and 1989 (~13 000 m^3s^{-1}), February 1986 (~14 000 m^3s^{-1}), January 1996 and 1999 (~15 000 m^3s^{-1}).

The average (for the 1990s) residence time of water within the estuary was estimated to be 35 and 40 days for the surface and bottom layers, respectively (Q_V = 26 000 m^3 s^{-1}; end-member salinities= 0.3 and 29 psu; surface and bottom salinities= 15 and 27 psu, respectively).

The relationship between wind stress and the depth of the mixed layer (defined by the depth of the halocline) was studied during two consecutive cruises in the estuary (May–June 1981; Q_V ~20 000 m^3 s^{-1}). Persistent moderate winds (2–7 m s^{-1}) were not able to break the vertical stratification for several weeks (see Fig. 4).

Nutrient dynamics

Tidal River

Nitrogen (DIN) and phosphorus (DIP) loads from Buenos Aires city untreated sewage were estimated to be about 25% of the total load entering the upper region in the 1980s (Pizarro & Orlando, *op. cit.*). Nagy (2000) estimated an average input of 3.5 × 10^6 mol DIP d^{-1} and 64 10^6 mol DIN d^{-1}. Typical concentrations of nitrate (25–40 μM), phosphate (0.5–2 μM) and ammonium (10–20 μM, see Fig. 5), and the N:P ratio (>25) were relatively high. The fraction of DIP adsorbed to suspended particulate matter (SPM) may be greater than the dissolved one (Pizarro & Orlando, *op. cit.*) and the N:P ratio decreased to <15 at the turbidity maximum (<0.5 psu) due to the decrease of nitrate to 20–30 μM and of ammonium to 2–5 μM.

During the El Niño event in 1983 (Q_V= ~40 000 m^3 s^{-1} in August), both the nitrate load and the N:P ratio increased ~75% (Nagy et al., 2001), whereas ammonium relative proportioning among DIN species decreased to < 10%.

The short-term variability of both nutrients and suspended matter was studied over 36 h at a fixed-point measurement at the turbidity maximum in November 1989 (Q_V = ~26 000 m^3 s^{-1}; surface salinity= 0–1 psu and bottom salinity= 1–5 psu). The tide-height continuously decreased (~0.7 m) because of a wind-induced water discharge episode, which allowed rapid water change and suspended sediment resuspension to be observed. An inverse relationship between DIP and SPM as well as a slight inverse relationship with tide-height was found; dissolved nitrate seemed to increase slightly with SPM, whereas DIP showed an opposite trend (see Fig. 6).

Silicate (DSi) concentrations typically varied from 140 to 220 μM (average = 188 μM; Pizarro & Orlando, *op. cit.*; Nagy & Blanco, 1987).

The range of oxygen saturation averages for the cruises conducted in the tidal river between 1981 and 1987 was 70–105% (average= 87%), and never dropped below 60%.

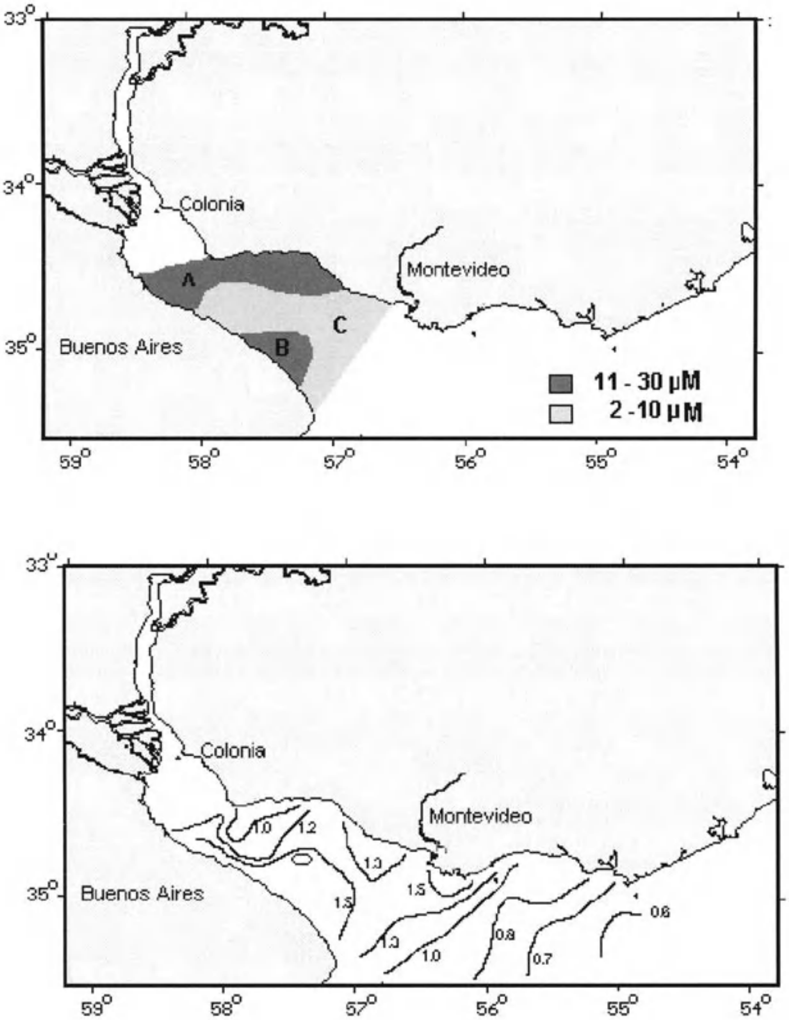

Figure 5. Average concentrations (μM) of ammonium in the tidal river (top) and phosphate (bottom) in the whole system (1981–1987). Modified from CARP (1990).

Table 3. Average nutrient concentrations (1981–1987). Freshwater end members for the tidal river (FW1) and estuary (FW2), system mean, and ocean end member. Modified from Nagy (2000)

Property	FW 1	FW 2	System	Ocean
Salinity (psu)	0.1	0.3	15.0	29.0
DIP (μM)	1.5	1.6	0.8	0.5
DIN (μM)	45.0	29.0	7.0	3.0
DSi (μM)	188.0	176.0	80.0	21.0

Frontal zone

The typical freshwater end member for NO_3^- was 20–29 μM. Nagy (*op. cit.*) estimated 29 μM DIN as the average input to the frontal zone (as shown in Table 3). Long-term averages of NO_3^- evolved along the salinity gradient in a non-conservative manner (see Fig. 7), with a sink estimated to be up to 60% on average, or ~75% during productive events (Nagy, *op. cit.*).

Internal sources were related to anthropogenic organic matter, resuspension from the turbidity maximum (see Fig. 6b), and benthic flux. The typical freshwater end member for PO_4^{3-} was ~1.5–1.6 μM. High values of phosphate, i.e. 3–5 μMwere exceptionally found in the bottom layer. The average freshwater end member for DSi was 176 μM (Nagy, *op. cit.*), similar to that reported by Meybeck (1981) for the average discharge of 62% of the total world runoff.

Outer zone

Prevailing concentrations of nitrate (2–10 μM), ammonium (1–3 μM), and phosphate (0.5–1.2 μM) in

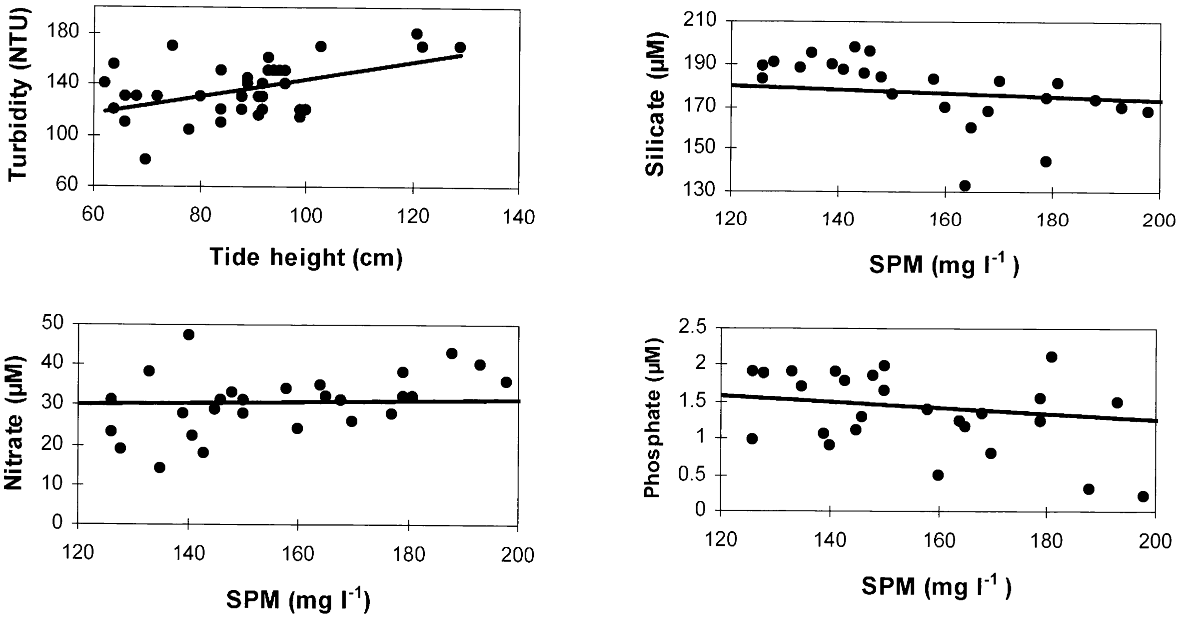

Figure 6. Relationships between: tide-height (cm) and turbidity (NTU); suspended matter (mg l^{-1}) and nitrate; suspended matter and silicate (µM); suspended matter and phosphate during a fixed-point measuerment at the estuarine turbidity maximum (November 1989).

the outer zone were typical of non impacted systems. Ammonium concentration and relative proportioning among DIN species generally increase along the salinity gradient, exceeding those of NO_3^- at salinities greater than 15 psu, especially during blooms, when NO_3^- may be depleted.

The typical mixing profile of phosphate has a slight decreasing trend, never falling below 0.1 µM, and the average is close to 1 µM. At low and intermediate salinities, some sink was observed, but less pronounced than for nitrate, whereas at higher salinities (>20 psu) phosphate tends to become steady. Phosphate concentrations in the bottom layer are generally higher than 0.5 µM (average= 1.05 µM and average salinity ~26 psu; as shown in Table 4). This is high in comparison with the reported coastal ocean end member for phosphates (0.5–0.6 µM at ~29 psu; Hubold, 1980a,b; Fillmann, 1990; Nagy, *op. cit.*).

Although there was some variability in DSi concentrations along the salinity gradient, it generally decreased seaward in a conservative manner (see Fig. 7).

Stoichiometric calculations

Average oxygen saturation in the bottom layer was <70% (AOU ~180 µM). Generally, both nitrate and phosphate increased with oxygen utilization (as shown in Table 4). The DIN:DIP ratio estimated release rate from the benthic environment fluctuated somewhat but remained below the classic Redfield ratio (average ~6, see Fig. 8 and Table 4), suggesting preferential release of DIP over DIN. Preformed concentrations were estimated to be 0.5 µM for phosphate and 1.0 µM for nitrate, suggesting that N:P ratio of benthic flux was actually greater, somewhere between 7 and 10. The oxygen-DIP relationship (see Fig. 8) followed the Redfield ratio, probably 10–25% below it, whereas the oxygen-nitrate relationship was about 40–60% below the expected ratio (see Fig. 8). Nagy (*op. cit.*) estimated the net ecosystem metabolism (7 mol m^{-2} yr^{-1}) and denitrification (−1.2 mol m^{-2} yr^{-1}) for the frontal zone from Redfield stoichiometry, and DIP and DIN sink. During the El Niño event (August 1983) nitrate concentration was >12 µM in the estuary in the bottom layer (as shown in Table 4) because of the increase of river flow rather than *in situ* mineralisation of organic matter (Nagy et al., 2000).

Phytoplankton biomass and nutrient depletion

Phytoplankton pigments have been measured at coastal stations close to cities and in open waters (as shown in Table 5). Excluding cyanobacteria and flagellate blooms, chlorophyll *a* concentrations varied from 0.05 to 15 µg l^{-1} (1981–1987 period). Typical values were below 10 µg chl. *a* l^{-1}, with a mode within the range 2–4 µg chl. a l^{-1}.

Table 4. Average chemical values for the bottom layer of the Rio de la Plata estuary (1981–1985). Salinity (psu), apparent oxygen utilisation (AOU, ml l^{-1}), nitrate, phosphate, dissolved inorganic nitrogen (μM), and total river flow (m^3 s^{-1})

Cruise	Salinity	AOU	O_2 saturation	Nitrate	DIN	Phosphate	N:P	River flow
1981–03	25.3	0.2	98	0.4	3.2	0.5	6.6	25 000
1981–05	26.3	3.2	43	6.2	8.0	1.8	6.2	21 000
1981–06	27	4.8	13	8.6	10.6	1.7	6.2	20 000
1981–09	26.3	0.8	88	1.5	3.1	0.7	4.4	16 000
1982–03	28.7	1.7	66	3.0	6.6	1.1	6.3	24 000
1982–07	29.9	2.3	64	2.4	12.4	1.3	9.5	29000
1982–09	25.7	2.7	59	2.3	2.7	0.8	3.4	31 000
1982–11	27.2	2.4	58	5.3	5.4	0.9	6.0	33 000
1983–08	26.8	1.8	77	12.8	14.7	1.1	13.0	40 000
1983–09	29.2	1.0	84	1.7	5	0.7	7.0	30 000
1984–06	22.9	4.4	20	10.4	12	1.9	6.3	31 000
1985–04	28.4	1.4	75	3.3	5.5	1.1	5.0	30 000
1985–10	25.8	1.3	79	3.2	4.4	0.4	11.0	25 000
Average	26.7	2.0	66	4.6	7.0	1.1	6.4	27 000

Table 5. Chlorophyll *a* concentrations (μg chl. *a* l^{-1}) in the Rio de la Plata tidal river and estuarine open waters. Mean, typical (mode), typical maximum (annual cycle, 1981–1987), and maximum values (November 1999) are reported

Chlorophyll-*a* ($\mu g\ l^{-1}$)	Tidal river	Frontal zone	Canal oriental	Marine zone
Mean value	1.0	2.0	2.0	2.0
Typical range	1.0–2.0	2.0–4.0	2.0–4.0	1.0-2.0
Typical maximum	15.0	15.0	12.0	10.0
Maximum value	28.0	38.0	22.0	unknown

Table 6. Susceptibility and level of expression of eutrophic conditions for each study site (Low= L; Moderate= m; High= H). Coastal point-source plumes are not considered. Low capacity to dilute (D) and/or flush (F) nutrients determines high susceptibility (S). Influencing factors are Dissolved Inorganic Nitrogen (N) and Phosphate (P). Primary symptom (PS) is chl. *a* (C), and secondary symptoms (SS) are low dissolved oxygen (O), and harmful blooms (B)

	Susceptibility			Influencing factors		Primary symptom	Secondary symptoms	
Study site	D	F	S	N	P	C	O	B
Tidal River	M	L	M	M	H	L	L	M
Frontal Zone	L	L	H	M	H	H	M	L
Canal Oriental	L	L	H	L	M	H	H	M
Outer Zone	L	L	H	L	M	L	M	L

Eutrophic values of algal biomass have been found in open waters in the Canal Oriental in November 1999 (up to 38 μg chl. *a* l^{-1}; see Figure 9), associated with very low River Uruguay flow (~1500 m^3 s^{-1}) and upward displacement of the front. In this cruise nutrient sink (see Fig. 9) reached ~65% for silicate, and mostly occurred at low salinities (<6 psu) in stratified, oxygen saturated (>120%) waters.

Cyanobacteria and flagellate blooms

Most of the extensive blooms were dominated by micro flagellates in the tidal river, micro flagellates and diatoms in the frontal zone, and centric diatoms and dinoflagellates in the Canal Oriental and outer region. In the tidal river, it was possible to find extensive blooms of cyanobacteria (*Microcystis aeruginosa*) in spring and summer, which have originated since 1980 in the Salto Grande dam reservoir, and reaching the Rio de la Plata through the River Uruguay discharge (Gómez-Erache & De León, *op. cit.*). The first registration of a toxic bloom of this cyanobacteria was reported in February 1999 close to the River Uruguay debouchement (De León & Yunes, 2001). Nevertheless there was no evidence of N-fixation.

Successive episodes of coastal mussel toxicity have been reported for the estuary since 1980 in Uruguay (Méndez, 1993). Periodic plankton studies show that toxic outbreaks of *Gymnodinium catenatum* and *Alexandrium tamarense* occurred in summer and early spring, respectively. Méndez et al. (1996) repor-

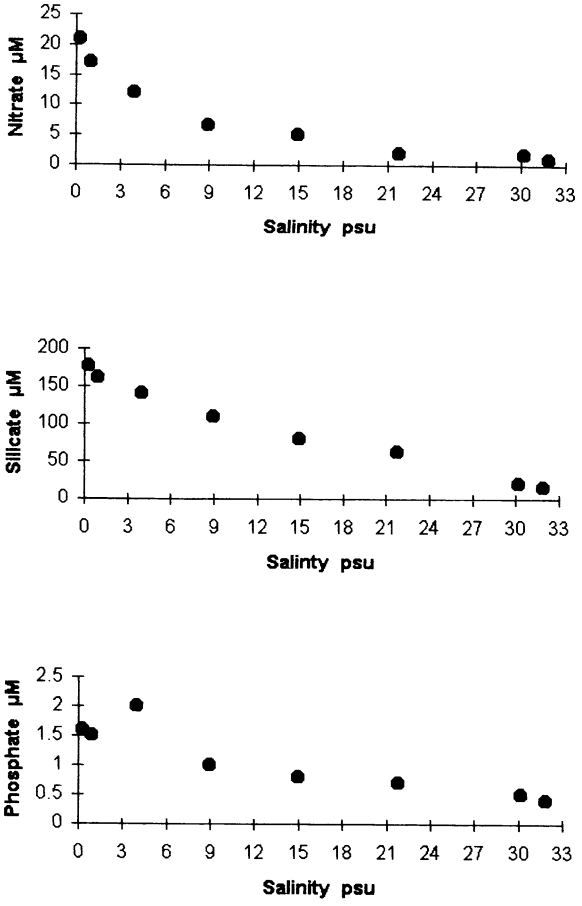

Figure 7. Relationships between average (1981–1987) salinity (psu) and nutrients (µM) in the surface layer: salinity vs nitrate; salinity vs silicate; salinity vs phosphate.

ted two strong toxic blooms in September 1991 (34 500 cells l^{-1}) and September 1993 (10 000 cells l^{-1}), and a moderate one in September 1992 (1000 cells l^{-1}). The authors attributed the blooms to two main environmental causes: an increment in the strength and northward displacement of the Malvinas current, causing the temperature range (11–15 °C) adequate for the development of *A. tamarense* blooms in the early spring, and a reduction of river discharge (the authors only considered the River Paraná discharge as freshwater inflow) with the consequent inflow of marine water (26–33 psu).

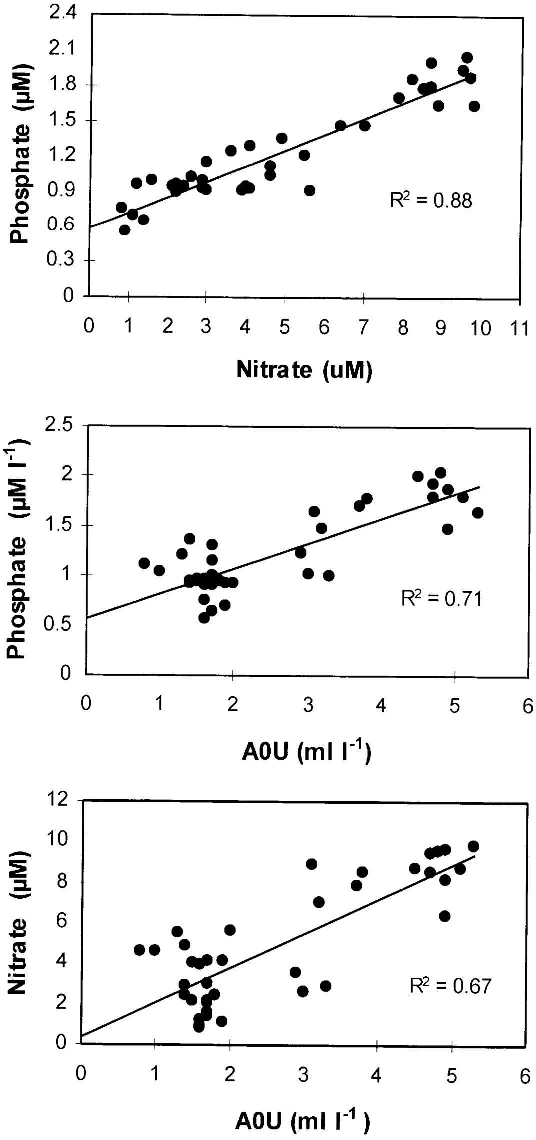

Figure 8. Relationships between Nitrate-Phosphate (top), AOU-Phosphate (middle), and AOU-Nitrate (bottom) in the bottom layer (Canal Oriental and marine zone) in June 1981. Phosphate and nitrate preformed values are ~0.5 and ~1, µM respectively. Apparent Oxygen Utilization (oxygen anomaly) = AOU (ml l^{-1}).

Eutrophication assessment

The estuarine susceptibility or 'capacity to retaining nutrients' and the eutrophic conditions were characterized as low, moderate and high (as shown in Table 6). Susceptibility is assumed to be directly related to estuarine stratification and volume, and inversely related to river flow and tidal range. The symptoms of eutrophication, i.e. chlorophyll *a* and dissolved oxygen concentrations, are determined from their concentrations, extent and frequency.

The tidal river is moderately susceptible and the estuary is highly susceptible, whereas the overall eutrophic conditions are moderate. Chlorophyll *a* and oxygen stress was retained as the descriptors of primary and secondary symptoms, respectively.

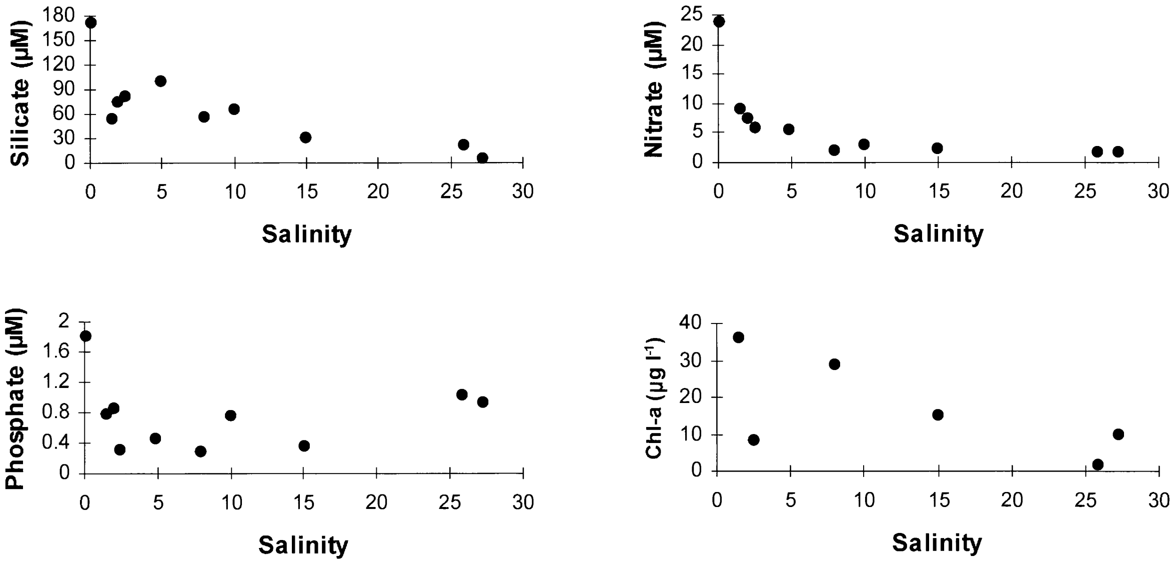

Figure 9. Mixing diagrams during a cruise conducted along the Canal Oriental in November 1999 (strong La Niña conditions): salinity vs chlorophyll *a*, salinity vs silicate; salinity vs nitrate; salinity vs phosphate.

Discussion

Nutrient dynamics

Tidal river and turbidity maximum

The downward decrease of DIN concentrations (from >40 μM to <30 μM), especially ammonium, within light limited waters suggests that both dilution of the polluted plume of Buenos Aires city (~25% of total load) and some denitrification occurs. Probably, some ammonium is adsorbed into SPM. During El Niño (i.e. August 1983; QV ~40 000 m^3 s^{-1}) the diffuse load of nitrate may have led to eutrophic concentrations (i.e. >70 μM) and high N:P ratio (>40).

Smith (1997) observed some discrepancy between the estimated inputs by Pizarro & Orlando (*op. cit.*) and phosphate concentrations in the tidal river. The high particulate DIP concentrations and some inverse SPM-DIP relationships found in some cruises and at the fixed-point measurements at the turbidity maximum (November 1989) show consistent relationships between tide-height, turbidity, phosphate and nitrate. An apparent equilibrium is reached within the ranges 100–150 mg l^{-1} and 0.8–1 μM for SPM and phosphate, respectively, suggesting the existence of interactions with suspended particulate matter, long since reported in the literature (Pomeroy et al., 1965; Froelich, 1989; Eyre, 1994). For phosphate a negative trend appears which could, in fact, be interpreted as a phosphate adsorption onto sediments. The adsorbed DIP fraction is thought to act as a buffer (Froelich, *op. cit.*; De Jonge & Villerius, 1989). Dissolved nitrate increasing trends may be attributed to both short-term river input variability and mixing with bottom enriched waters.

Data from cruises in the tidal river and the fixed-point measurements (see Fig. 6) suggest that in freshwater (<0.5 psu) the concentrations of both SPM (100–140 mg l^{-1}) and DSi (170–180 μM) are linked. This seems to be related to both short-term increase of water discharge induced by wind, and resuspension of the suspended sediment turbidity maximum.

Estuary

Riverine discharge usually remains at the same magnitude on the weekly-monthly timescale; hence the freshwater end member is assumed to remain constant in time over the residence time (~40 days) of the water in the estuary.

Mixing diagrams for nitrate show that this nutrient is strongly removed in the estuary within the salinity range 0.2 and 18 (see Fig. 7). Nagy (*op. cit.*), based on long-term average values, concluded that DIN removal is 60% of total input, and a half could be due to denitrification (−1.3 mol m^{-2} y^{-1}). In November 1999, during La Niña (Q_U<2000 m^3 s^{-1}), nutrient sink along the Canal Oriental, mostly via assimilation, is estimated to have been about 80% for nitrate, 65% for silica, and 60% for phosphate (see Fig. 9). The degree to which nutrient modification proceeds is largely controlled by estuarine flushing time (Peterson et al., 1985; Church, 1986; Balls, 1994), which is primarily regulated by river discharge (Eyre & Twigg, 1997).

The balance of dissolved phosphate source-sink suggests that at least ~30–50% is added/removed at the turbidity maximum, probably more than the biological removal and sewage input in the frontal zone. High phosphate concentrations (i.e. >3 μM) sometimes found in the bottom estuarine layer close to the turbidity maximum (<10 psu) seem to have originated from desorption, benthic flux, and anthropogenic flux sources. Long-term average steady DIP concentrations at high salinities (>20 psu) may be sustained by desorption and benthic flux.

The level of stratification in the water column is crucial in controlling the intensity of vertical mixing and hence the vertical fluxes of water properties such as heat, salt, momentum, and the biogenic elements (Simpson et al., 1990). The breakdown of stratification, and the resuspension and shift of location of the suspended sediment turbidity maximum, is a key factor to understand nutrient behavior in the lower tidal river and estuarine turbidity zone, as well as some apparent discrepancies, i.e. the change of the N:P ratio and the increase of DIP and DSi values sometimes observed in the frontal zone.

The long-term average mixing diagrams for silicate show maximum removal within the salinity range 1–18 with different phytoplankton composition, and nutrient and light availability. Removal is estimated to vary from 20% (see Fig 7) for average conditions up to 70% for highly productive events (see Fig. 9).

Stoichiometry

The N:P ratio of the inorganic nutrients shown for suboxic bottom waters (~7; as shown in Table 4), once corrected for typical preformed phosphate and nitrate values is ~8–10. Nagy et al. (2000) found that both AOU and river flow explain most of the variability of nitrate and phosphate concentrations within the salinity range 25–30 psu. Low N:P ratios are in part attributable to nitrogen loss by denitrification (Seitzinger, 1988). The potential limitation (N:P <10, Dortch & Witledge, 1992) is also imposed by benthic release. Stratified waters in the lower Canal Oriental and outer zone are limited by nitrogen (N:P = 1–4 at the surface layer). The difference found between average bottom layer phosphate concentrations (1.05 μM) and reported values for the adjacent shelf surface waters (0.5–0.6 μM), is consistent with the oxygen utilization (A0U= 180 μM) converted into phosphate (plus 0.5–0.6 μM) according to Redfield stoichiometry. Therefore, both phosphate and oxygen seem to be useful for stoichiometric calculations of net ecosystem metabolism in the Rio de la Plata, in spite of the existence of some surface processes which are also involved.

Nutrients, river discharge, and toxic blooms

Nutrient variability is frequently associated with changes in the adaptive advantages among populations with different nutrient requirements (Tilzer, 1989). In particular, silicate may be the controlling nutrient in shifting production from diatoms to flagellates (Officer & Ryther, 1980). In November 1999 (River Uruguay discharge <2000 $m^3 s^{-1}$) DSi concentrations in the lower Canal Oriental were below the limiting value of 5 μM suggested by Fisher et al. (1988) (see Fig. 9), as well as below the typical ocean end member (~21 μM, as shown in Table 3). This depletion of DSi is probably the result of significant use of recycled nitrogen by the bloom during low fresh inflow episodes, increased residence time of both water and plankton, and stratified conditions at the frontal zone.

The reduction of River Uruguay discharge (<2000 $m^3 s^{-1}$) in both September 1991 and 1993 seems to have controlled the strong *Alexandrium tamarense* toxic blooms reported by Méndez et al. (1996). This hypothesis is supported by two facts: in September 1992, when the discharge was 5200 $m^3 s^{-1}$, the bloom was moderate within the system, and the strength and northward displacement of Subtropical confluence was similar during the three bloom periods (September 1991, 1992 and 1993).

Phytoplankton biomass, nutrients and trophic state

Golterman & Oude (1991) stated that in comparing systems, chlorophyll *a* remains the only useful, practical and simple measure of trophic state (TS) ranking. Based on mean chlorophyll stock, both the tidal river and estuary are mesoeutrophic (Gómez-Erache et al., 2001a). The frontal zone is autotrophic in the long-term; nevertheless, during prolonged (weeks) stratification episodes, respiration below the halocline might become greater than surface production.

Overall susceptibility, influencing factors, and eutrophic conditions

The overall ranking of primary and secondary symptoms (chlorophyll *a*, and nuisance/toxic blooms and biological stress respectively) determines that the level of expression of eutrophic conditions is moderate (as shown in Table 6). However, natural susceptibility, present conditions and future outlook are different at each study site.

The tidal river is moderately susceptible and eutrophic (nutrification and occurrence of cyanobacteria blooms). However, as stated by Irigoïen & Castel (1997) for the Gironde estuary, the strong limitation by turbidity, as well as mixing by wind, allows the ecosystem to absorb large inputs of nutrients without developing severe symptoms of eutrophication.

The estuary is highly susceptible (highly stratified, large volume, low tidal range), and moderately eutrophied (moderate chlorophyll *a* concentrations, oxygen stress, harmful blooms). Only 14% of the highly eutrophied estuaries in the United States have high-level nitrogen input, whereas 57% exhibit a high susceptibility (Bricker et al., *op. cit.*). Within the frontal zone such processes like nutrient removal, *in situ* production of organic matter, and denitrification are estimated to be high (Nagy, *op. cit.*). This should reduce part of the eutrophication danger seaward. Nevertheless, if river flow and/or human impacts increase, denitrification sink should be lower with increasing anoxia (Nagy et al., 2001). Coastal ecosystems in which phytoplankton growth is frequently limited by nutrients (i.e. the marine zone) are highly sensitive to changes in nutrient loading (Cloern, 1999).

The Canal Oriental may be considered as a conduit channel. Deep channels are an effective mass conduit because of higher residual velocities (Lucas et al., 1999). Therefore, River Uruguay flow should act as a controlling factor on both phytoplankton blooms development and horizontal transport, besides the control it exerts on vertical stability. Stratification is favorable for high levels of *in situ* organic matter production, which in turn is oxidized below the halocline. If stratification lasts over several consecutive tidal cycles it is called ‘runaway stratification’ (Simpson et al., *op. cit.*). Because of its low tidal mixing, shallow mixed layer ($\sim$5 m depth), and depth (10–25 m), the estuary behaves as ‘runaway stratified’. In the Canal Oriental the occurrence of long episodes (weeks) of permanent 3b or 4 S-C type allows for the development of hypoxic conditions and the remineralisation of nutrients (i.e. May–June 1981). This subsystem is also susceptible to the development of symptoms of eutrophication even at the usually nutrient-depleted surface layer.

Within the Canal Oriental the pronounced reduction of freshwater inflow during La Niña (i.e. November 1999) and the consequent increase of flushing time, leads to eutrophic symptoms (i.e. >20 μg Chl. a l^{-1}) and changes in nutrient ratios. Finally, the findings presented here suggest that the physical structure of the water column and the energy available for mixing, as well as the trends in quantity and quality of freshwater and nutrient loads (especially nitrate) determine that eutrophic conditions should worsen a lot in the next decades.

The application of precautionary principles should not be delayed. Fortunately, awareness by financial and management institutions of the need for environmental planning and a clean up in the region is mounting (Gómez-Erache et al., 2001b).

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Baseline study of soft bottom benthic assemblages in the Bay of Santander (Gulf of Biscay)

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Key words: baseline, soft bottom benthos, sediments, organic pollutants, heavy metal

Abstract

The Bay of Santander constitutes one of the most important estuarine areas in northern Spain (Gulf of Biscay) which has been significantly stressed by sewage discharges. However, an improvement in the environmental quality of the bay is expected once the new sewer system comes into operation. The evaluation of such an evolution needs to be contrasted with the current situation, established in specific baseline studies. For this purpose, a field study was carried out on the soft bottom assemblages of the estuary in spring of 1998. At each station, the benthic fauna abundance, grain size, organic matter, carbon, nitrogen, phosphorus and heavy metals concentrations were analysed. The community structure and the physical and chemical characteristics of the sediment reflected the higher impacts both near the points of discharges and in the low water exchange zones, whereas those areas showing higher tidal flows presented an acceptable quality. High heavy metal concentrations for Cd, Hg, Pb and Zn indicated contamination of industrial sources in several areas of the Bay. This information allows to establish a reference situation in order to improve the design of a specific monitoring program for this area.

Introduction

Soft bottom benthic communities very often reflect the effects of pollution and have been successfully used to indicate anthropogenic impacts in estuarine ecosystems (Pearson & Rosenberg, 1978; Gray, 1979; Boesch & Rosenberg, 1981; Warwick et al., 1990). Thus, monitoring programs of environmental effects caused by sewage discharges usually concentrate on this component of the ecosystem (Warwick et al., 1990; Ferraro et al., 1991; Warwick, 1993; Otway, 1995). However, patterns of response to pollution in benthos depend on the resistance of each community and the nature and timing of the perturbation (Boesch & Rosenberg, 1981).

The Bay of Santander (N Spain, Gulf of Biscay) is an important economic, recreational and natural estuarine area, which has been significantly stressed by the continuous untreated discharges from urban and industrial sources. An improvement in the environmental quality of the estuarine ecosystem is expected once the new sewer system comes into operation. However, as shown in previous research (NRC, 1992), the short-term recovery in pelagic communities will contrast with longer time period required for benthic remediation. Furthermore, the ability and the time necessary for recovery of the latter will be different along the estuary, depending on the spatial distribution of the current continuous discharges, the environmental gradients (e.g. grain size, hydrodynamic features) and the resistance and resilience of the affected communities (Boesch & Rosenberg, 1981; Díaz-Castañeda et al., 1993; Díaz & Rosenberg, 1995; Bellan et al., 1999).

Hence, a monitoring program is required in order to test the effectiveness of the sanitation project and to know how the recovery process takes place along the estuary. The design of suitable monitoring programs should be based on the knowledge of the current situation (Hammerton et al., 1981). The usual statistical approaches for the detection of patterns at the community level combine both univariate and multivariate methods (Field et al., 1982; Clarke & Warwick, 1994). Therefore, baseline studies have been carried out in order to, first, recognise and characterize great-scale

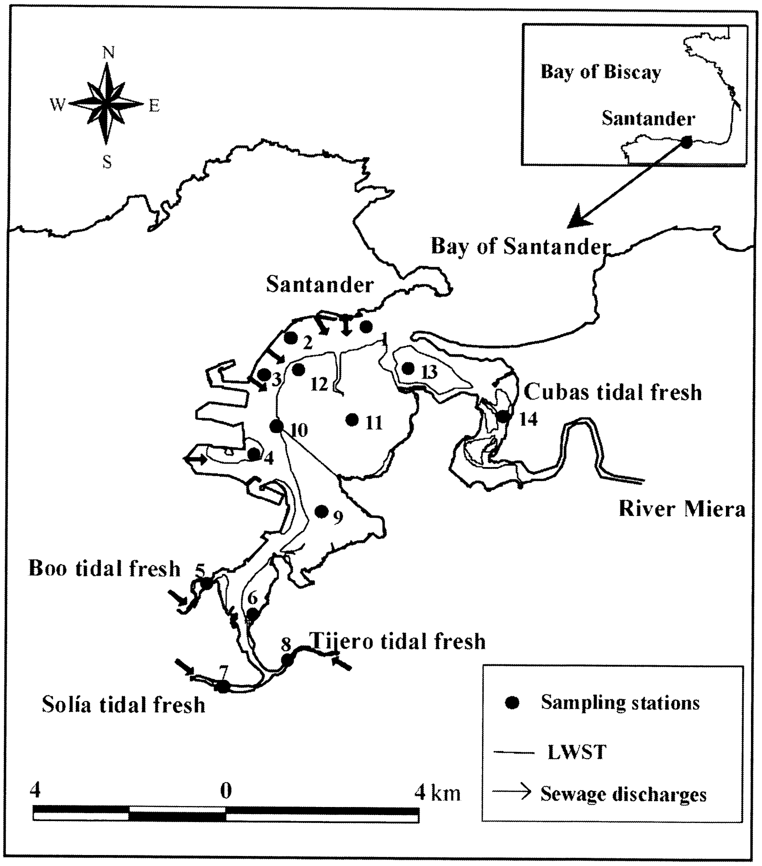

Figure 1. Location of sampling stations (1–14) within the study area, with indication of the low water spring tide (LWST) level (fine line).

areas, according to the specific faunal assemblages and, secondly, analyse their relationships with the ranges of natural and man induced conditions. Based on those results, we attempted to examine the sensitivity of different statistical analysis for the detection of changes in benthic communities, regarding their future application to the design of the monitoring program.

Study area

The Bay of Santander constitutes one of the most important estuarine areas in Northern Spain (Gulf of Biscay) (Fig. 1), with a total extension of 22.5 km^2, of which 65% are intertidal areas. Subtidal zones are dominated by shallow waters, with maximum depths of 10–12 m occurring along the navigation channel. Soft-bottoms dominate throughout the Bay. Hydrodynamic conditions are controlled by semidiurnal tidal regime and 3 m mean tidal range, interacting with variable freshwater inputs coming mainly from the river Miera through the Cubas area and, to a much lesser extent, from small streams through the Boo, Tijero and Solía tidal fresh areas (river inlets).

Materials and methods

A survey was carried out during the spring of 1998 (second half of May). Fourteen soft bottom stations were sampled, both in the subtidal (stations 1, 2, 3 and 10) and in the intertidal (stations 4–9 and 11–14) areas of the estuary (Fig. 1). Three replicated samples were collected in each station, two for analysis of the benthic macrofauna and one for physical and chemical characterization of sediments. At the intertidal stations, 2500 cm^2 and 15 cm deep surface sediments were collected by hand-digging (Eleftheriou & Holme, 1984). At the subtidal stations, 625 cm^2 and 15 cm deep, samples were obtained by scuba divers, using a suction sampler. Each replicate, both at the intertidal and the subtidal points, was sieved through

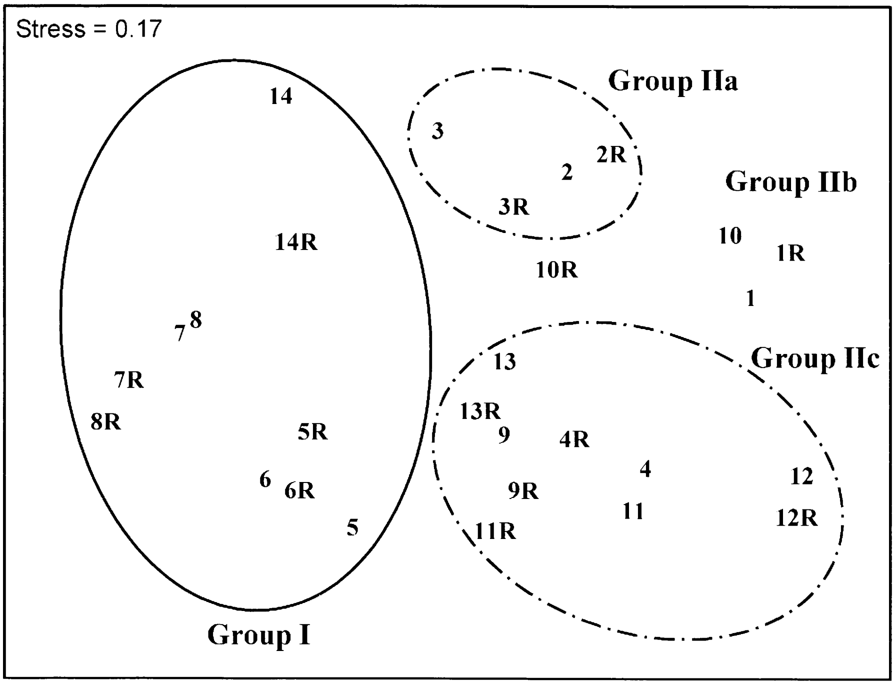

Figure 2. Multidimensional scaling ordination of samples for biomass data (R=replicate sample). The groups established from the cluster analysis are indicated (continuous line at 9% similarity level and discontinuous line at 23% similarity level).

a 0.5-mm mesh screen to separate macrofauna. All organisms retained on the screen were preserved in 5% formalin until sorting and identification of specimens.

In the whole set of sediment samples, grain size, redox potential, organic matter content and elemental analysis (C, N, P) were determined. Granulometry was determined by dry sieving following the Wentworth scale. The redox potential was measured with an ORION 250A platinum electrode. The organic matter was estimated from dried sediments as loss on ignition. Total carbon and total nitrogen were determined using a CNH analyser and total phosphorus by acid digestion. Heavy metal concentrations (Hg, Al, Cd, Cu, Cr, Ni, Pb and Zn) were measured on the <63 μm fraction by flame atomic absorption. This variable was only determined in 10 of the 14 stations studied (2–10 and 12), selected according to the probability of affection by industrial sources. Species richness (number of taxa), abundance (number of individuals m^{-2}), biomass (g dry weight m^{-2}), numerical diversity (Shannon–Weaver, 1963, bits ind^{-1}) and evenness (Pielou, 1969), were calculated for each sample after identification to the lowest possible taxon, usually species. The Abundance-Biomass Comparison method (ABC plots) was also used in order to determine levels of disturbance on those samples (Warwick, 1986).

Hierarchical agglomerative clustering, with UPGMA as linkage option, and ordination by Multidimensional Scaling (Kruskal & Wish, 1978) were performed on fourth-root transformed macrofauna abundance and biomass data, using the Bray-Curtis similarity. Multivariate BIOENV procedure (Clarke & Ainsworth, 1993) were used to explore the potential relationships between the abiotic features of sediments and the similarity patterns among biological samples. Mean annual salinity values, obtained in previous studies (Moreno-Ventas, 1998), were included in this analysis. Multivariate analyses were performed using PRIMER software (Plymouth Routines in Multivariate Ecological Research). Combined interpretation of the statistical analysis will be used for the recognition of homogeneous benthic areas according to environmental gradients.

Results

Ninety one species of macrofauna are registered in the 28 samples analysed, with ranges of richness between 3 and 21 species per sample and 8–9 species on average, in both the intertidal and the subtidal zones. The macrobenthos is characterised by a great dominance of polychaetes followed by molluscs, mainly bivalves. Many taxa colonise simultaneously the intertidal and subtidal habitats, although relative abundance of macrofauna species differs in both areas. Thus, at the subtidal areas, *Hinia reticulata*, *Euclymene oerstedi*, *Notomastus latericius* and *Polydora ciliata* are the most frequent species (88%, 88%, 63% and 50% of presence in samples, respectively), and at the intertidal, *Scrobicularia plana* (55%), *Cirriformia tentaculata* (50%), *Nephtys hombergii* (50%), *Cerastoderma edule* (50%), *H. reticulata* (45%) and *Dosinia exoleta* (45%).

Cluster analysis based on biomass data established differences between stations located in the surroundings of tidal fresh areas (group I) and the other stations (group II), at a 9% similarity level. Furthermore, three subgroups can be defined in the latter, at 23% similarity level, corresponding to subtidal stations 2 and 3 (subgroup IIa), subtidal stations 1 and 10 (subgroup IIb), and intertidal flats located mostly in the eastern part of the estuary (subgroup IIc). Using abundance data (number of individuals), the plot does not change noteworthy, except for station 12 which is clearly separated from the others samples of group II. The MDS ordination shows a spatial distribution of samples in agreement with the dendrogram (Fig. 2). Thus, samples from fresh tidal areas appear on one side of the horizontal axis separated from the rest of samples, which are ordered from intertidal flats (subgroup IIc), at the bottom of the vertical axes, through subtidal stations 1 and 10 (subgroup IIb), to subtidal stations 2 and 3 (subgroup IIa). Samples from fresh tidal areas show a gradation of change from stations 5 and 6 (Boo-Pontejos) to station 14 (Cubas), with stations 7 and 8 (Solía and Tijero) located in an intermediate position of the vertical axis. Nevertheless, the great dispersion of the samples in this group should be noted.

The average values for the ecological indices calculated reveal clear differences between the community structure in the groups distinguished by multivariate analyses (Table 1). The highest values of species richness and diversity are found in the intertidal flats (subgroup IIc), whereas subtidal stations included in subgroup IIb and those located in the surroundings of tidal fresh areas (group I) present low values of both parameters. In the latter, the independent analysis of the three station groups separated by the MDS plot shows differences among the values for the ecological parameters. At the Boo-Pontejos area, richness and diversity yield intermediate values, whereas Solía, Tijero and Cubas tidal fresh areas attain the lowest values. Subtidal stations of subgroup IIa show high values of richness, similar to those found in the intertidal areas, although diversity was lower. The highest number of individuals was recorded in subtidal stations of subgroup IIa and the Boo-Pontejos area, and the lowest values were founded at the other subtidal stations and in the Cubas tidal fresh area. The latter also shows the lowest values of biomass, followed by subtidal stations of subgroup IIa. The highest biomass values were recorded in the intertidal flats.

Figure 3 shows a selection of ABC plots representative of the environmental conditions observed in the previously defined areas. The plots of subtidal stations of subgroup IIa are characteristic of disturbed environments (cf. Fig. 3a), whereas those from Boo-Pontejos area (cf. Fig. 3e) and the Cubas tidal fresh area (cf. Fig. 3f) reflect the existence of a moderately disturbed community. Conversely, the ABC curves of the other two subtidal stations (cf. Fig. 3b) and intertidal flats (cf. Fig. 3c) show a pattern typical of undisturbed areas, except for stations 4 and 12, where a moderate degree of stress can be deduced (cf. Fig. 3d). The low number of species collected in the inner stations of the estuary (Solía and Tijero tidal freshs) is itself indicative of altered communities, and restricts the reliability of that analysis.

The characteristics of sediments along the estuary are shown in Table 2. The sampling stations are constituted mostly of sandy sediments, with percentages of the silt fraction below 5%, except in stations 3, 7 and 10. Maximum values of organic matter, total carbon, total nitrogen and total phosphorus occur in the subtidal (stations 1, 2, 3 and 10) and in the inner part of the estuary (stations 5, 6, 7, 8 and 14), whereas the concentrations are low in the intertidal flats (stations 4, 9, 11, 12 and 13). The distribution of heavy metals concentrations is similar to that described for organic contamination, the maximum values corresponding to the samples from the Solía and Tijero tidal freshs areas. The values of cadmium and, to a lesser extent, of zinc, lead and mercury registered in some stations indicate pollution of industrial sources. It is worth pointing out the very high Cd concentrations presented in the Solía and Tijero tidal fresh areas and the high concentrations of Hg in the subtidal station 2.

The highest single correlation established by the BIOENV analysis occurs with salinity and the concentration of copper (0.53), followed by that of total phosphorus (0.48) (Table 3). The combination of two or more variables does not increase significantly or degrade this correlation.

Table 1. Mean values of richness (number of species per sample), abundance (number of individuals m^{-2}), biomass (g dry weight m^{-2}), diversity (H, bits individuals^{-1}) and evenness, in the different areas distinguished by multivariate analyses (standard deviation is indicated in brackets)

		Richness	Abundance	Biomass	Diversity	Evenness
Group I	Global	5.60 (2.55)	376 (411)	52 (40)	1.31 (0.59)	0.53 (0.15)
	Boo-Pontejos (stations 5, 6)	8.00 (2.16)	574 (611)	76 (41)	1.78 (0.36)	0.60 (0.09)
	Solía-Tijero (stations 7, 8)	3.50 (0.58)	320 (159)	47 (34)	0.69 (0.15)	0.38 (0.04)
	Cubas (station 14)	5.00 (1.41)	92 (6)	11 (7)	1.60 (0.22)	0.70 (0.03)
Group II	Subgroup IIa (stations 2, 3)	12.00 (8.2)	554 (557)	18 (17)	1.68 (0.72)	0.55 (0.24)
	Subgroup IIb (stations 1, 10)	5.75 (1.50)	87 (73)	32 (24)	1.99 (0.43)	0.79 (0.10)
	Subgroup IIc (intertidal flats)	13.90 (3.60)	288 (118)	105 (54)	2.78 (0.27)	0.75 (0.10)

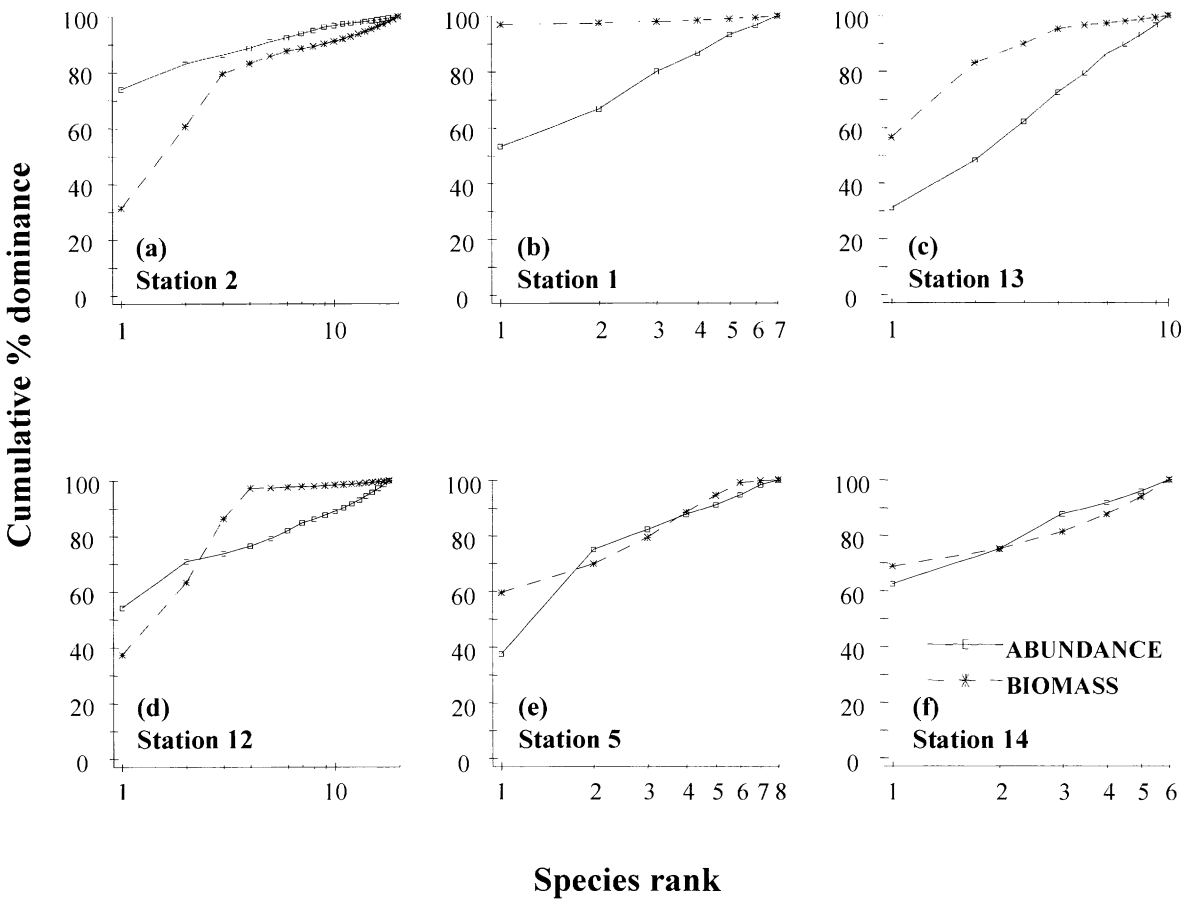

Figure 3. Selection of ABC plots of samples representatives of different conditions within the estuary.

Table 2. Granulometric distribution (% per fraction), chemical characteristics (mV, % and mg kg^{-1}, respectively) and heavy metal concentrations (mg kg^{-1}), of sediments in each sampling station (–=samples not analysed)

	Sampling stations													
	1	2	3	4	5	6	7	8	9	10	11	12	13	14
Very coarse sand (%)	9.0	35.2	25.5	4.4	43.3	43.9	37.4	51.3	19.2	35.3	8.8	0.6	1.6	32.2
Coarse sand (%)	5.8	10.3	7.0	1.6	8.2	17.4	11.8	18.6	6.0	8.8	7.6	1.9	16.2	26.8
Medium sand (%)	56.3	32.5	22.9	82.9	19.8	24.5	12.6	21.4	46.1	15.4	82.3	91.1	77.2	29.3
Fine sand (%)	15.4	13.2	20.6	7.7	16.8	8.2	18.2	4.8	16.2	20.3	0.8	4.6	3.4	7.3
Very fine sand (%)	11.8	7.2	18.4	3.4	10.0	5.2	9.6	3.4	9.6	11.2	0.4	1.7	1.4	4.2
Silt (%)	1.7	1.6	5.6	0.0	1.9	0.9	10.4	0.6	2.8	9.0	0.2	0.1	0.2	0.2
Redox (mv)	−125	−187	−190	−52	−212	−243	−168	−183	−76	−324	−126	−81	−124	−124
Organic matter (%)	9.1	8.19	6.83	1.31	5.29	8.93	7.84	11	2.84	10.63	1.19	1.52	1.45	5.82
Carbon (%)	4.81	4.12	3.51	0.82	2.64	4.61	4.12	7.03	1.39	5.92	0.85	0.74	0.77	3.12
Nitrogen (%)	0.47	0.58	0.41	0.33	0.64	0.81	0.59	1.03	0.4	0.66	0.18	0.29	0.19	0.35
Phosphorus (mg/kg)	551	514	600	112	357	922	1251	1076	184	654	117	144	182	727
Hg (mg/kg)	–	1.5	0.6	0.1	0.5	0.8	0.5	0.6	0.3	0.8	–	0.08	–	–
Al (mg/kg)	–	20 254	21 855	2050	22 902	28 342	36 017	43 548	11 478	29 282	–	2193	–	–
Cd (mg/kg)	–	5	5	<1	5	12	16	23	3	7	–	<1	–	–
Cu (mg/kg)	–	27	27	3	26	71	83	85	11	31	–	2	–	–
Cr (mg/kg)	–	32	32	4	41	111	243	153	19	47	–	5	–	–
Ni (mg/kg)	–	13	14	<4	14	28	43	51	6	17	–	<4	–	–
Pb (mg/kg)	–	108	76	14	71	116	114	118	39	112	–	15	–	–
Zn (mg/kg)	–	436	281	27	252	543	650	535	136	425	–	19	–	

Discussion

Combined use of univariate and multivariate statistical analyses has allowed different regions within the estuary to be recognised according to their faunal assemblages. Furthermore, this spatial distribution of the soft bottom communities can be related to both natural environmental gradients and levels of pollutants.

The differentiation established by multivariate analyses between group I and II can be related to the tidal/fresh water influence and the hydrodynamic conditions, reflecting a classical estuarine gradient. The gradations of community composition according to these factors appear quite well defined in the MDS. Thus, the sample sites located in the inner part of the estuary, with a great continental influence (mean salinity values between 15 and 27.2%; Moreno-Ventas, 1998), and low water exchange and current velocity (Revilla et al., 1998), appear at one end of the horizontal gradient. In contrast, stations 1, 10 and 12, located in the outer part or near the navigational channel, and therefore with a high tidal influence (mean salinity values between 32.9 and 33.8%; Moreno-Ventas, 1998) and higher velocity of tidal currents (Sofialidis & Prinos, 1998), appear on the opposite end.

Otherwise, the gradation established in the vertical axis of MDS within each group can be explained by differential affections of contamination. This argument is supported both in registered values of organic contaminants and heavy metal concentrations in sediments, ecological indices and ABC plots. Therefore, samples of group II are ordered, according to level of contamination, from intertidal flats, with low values of organic compounds and heavy metal, to subtidal stations with moderate to high concentrations of pollutants. According to this pattern, the former show high values of richness, diversity and evenness and ABC plots characteristics of unperturbed environments, except for stations 4 and 12. Both of these are located very near the channel of the estuary, where tidal currents can generate an unsteady environment (Sofialidis & Prinos, 1998). This physical disturbance can produce confounding effects with those of pollut-

Table 3. Summary of results from BIOENV analysis, corresponding to single variables (ρ=Spearman rank correlation)

Variable	ρ
Salinity	0.53
Cu	0.53
P	0.48
Ni	0.47
Cd	0.42
Cr	0.42
Zn	0.42
Al	0.40
Very coarse sand	0.39
Hg	0.39
Coarse sand	0.38
Medium sand	0.33
Pb	0.25
C	0.25
Organic matter	0.22
N	0.21
Silt	0.12
Very find sand	0.09
REDOX	0.09
Fine sand	0.06

ants (Turner et al., 1995). Also, some authors suggest that the ABC plot showing the 'perturbed' or 'moderately perturbed' configuration should be interpreted cautiously if the taxa present in high abundance are not polychaetes (Dauer et al., 1993; Warwick & Clarke, 1994; Smith, 1994). This is the case of communities analysed in this study.

Otherwise, although the sediment quality is similar in all subtidal samples, ABC plots may reflect differences in the degree of contamination. Thus, subgroup IIa (stations 2 and 3) shows an abundance-biomass distribution characteristic of polluted areas, whereas those included in subgroup IIb (stations 1 and 10) correspond to non-polluted conditions. Proximity of stations 2 and 3 to the main discharge points implies a more continuous and more direct exposition to pollutants, increasing the probability of occurrence of episodic deleterious effects, as those related to hypoxia events (Díaz & Rosenberg, 1995).

Ecological distances within fresh tidal areas in MDS again correspond to differences in environmental conditions, from Solía-Tijero (stations 7, 8), with minimum values of richness and diversity and very high concentrations of organic and industrial pollutants, to Boo-Tijero area (stations 5, 6), with moderate degree of contamination. Nonetheless, Cubas tidal fresh area (station 14) does not follow this pattern, even though its organic matter and phosphorus values are quite high. This may be due, on the one hand, to the higher influence of the continental input through the River Miera, and on the other hand, to the absence of industrial discharges to this area.

The results of BIOENV support, in part, these patterns in relation to estuarine gradients and increased levels of pollutants. Higher correlations correspond to salinity, reflecting the differential influence of tide and fresh water inputs, copper, indicative of contamination of industrial sources, and phosphorus, related to urban discharges. However, the low correlation values can be explained because of confounding effects with a range of environmental variables, as depth or sediment structure, that tend to obscure any relationships between faunal composition and levels of pollutants (Ferraro et al., 1991; Underwood, 1994; Chapman et al., 1995). Correlation with grain size is quite low, although this is recognised as a major factor influencing benthic community structure. However, the tendency of silty sediments to inter-grain cohesion may have produced confusing results.

Consequently, the identified areas within the Bay of Santander constitute a basic approach to the differentiation of estuarine environments to be considered in future studies. However, the great variability in physical, chemical and, overall, biological features along the estuary, even for replicated samples, advise the assessment of the spatial variability, at a smaller scale. In addition, temporal variability in those areas has to be taken into account before designing the appropriate sampling schedule.

The complementary use of univariate index or graphical/distributional methods helps to determine the level of stress and to characterize community structure in each area (Simboura et al., 1995). In addition, multivariate techniques used have provided a comprehensive pattern in faunal changes related to the main environmental gradients. Nevertheless, the results obtained are not always in agreement with those expected by concentrations of pollutants. Thus, it is advisable to use a range of analytical methods in order to ensure the detection of changes in benthic communities in response to sediment or water quality (Dauer, 1993; Nicolaidou et al., 1993).

Conclusions

Three general areas can be established throughout the Bay: (i) fresh tidal areas in the inner parts of the estuary, with higher freshwater influence, lower water exchange and moderate to high levels of pollution; (ii) subtidal areas located near the most significant urban discharges points; (iii) intertidal areas on the eastern side of the bay which present benthic assemblages more characteristic of unperturbed environments. The quality of sediments reflects a moderate level of organic and heavy metal pollution in the subtidal stations and a high level in the inner part of the estuary.

Acknowledgements

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Nutrient (N, P, Si) fluxes between marine sediments and water column in coastal and open Adriatic

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Key words: benthic flux, nutrient, marine sediment, Adriatic Sea

Abstract

Nutrient benthic fluxes, as well as sediment phosphorus concentration at the open sea and coastal water stations of the Central and South Adriatic were studied during 1997–98. The fluxes were in the ranges: 0.16–2.67 mmol m^{-2} d^{-1} (silicate); −0.031–0.164 mmol m^{-2} d^{-1} (phosphate); −0.51–2.03 mmol m^{-2} d^{-1} (ammonia); and −1.32–1.62 mmol m^{-2} d^{-1} (nitrate + nitrite). Silicate flux showed a gradient from the coastal area to the open sea. Ammonia was the main nitrogen species in the flux at the estuary and bay stations, while the sum of nitrate and nitrite was predominant at the open sea stations. Relationships between phosphate and ammonia fluxes ($r = 0.699$, $p<0.01$) as well as phosphate and silicate ($r = 0.529$, $p<0.01$) were established.

Introduction

As a consequence of organic matter oxidation in the marine sediment (Froelich et al., 1979) fluxes of inorganic nutrients between the sediment and the overlying seawater are established through the sediment-water boundary layer (Santschi et al., 1983; Jørgensen & Revsbech, 1985; Sundby et al., 1992). The benthic nutrient fluxes are, in general, diffusion controlled, but in shallow areas they can be enhanced through infaunal activity (Tahey et al., 1994) or hydrodynamic pressure (Vanderborght et al., 1977). The importance of benthic fluxes as an important source of nutrients for the primary productivity was pointed out by numerous authors (e.g. Rowe et al., 1975; Hopkinson, 1987). The fluxes can be also enhanced due to increased nutrient input to a particular area (Berelson et al., 1998), but on a wider scale it was showed that through hydrographical circumstances the continental slopes (not the shelf areas) are the major deposition centres of organic matter and areas of intensive remineralisation (Christensen, 1989; Lohse et al., 1995; Walsh, 1991).

Hammond et al. (1984) and Giordani & Hammond (1985) studied nutrient fluxes in the Northern eutrophic Adriatic. Their results showed that the obtained benthic flux was comparable to the input of nutrient from the Po River, while further investigations (Giordani et al., 1992) showed that a macrofauna plays important role in this area. In addition, Banzon & Hopkins (1995) investigated the estuary of the Po River, the entire Italian coast and the Jabuka Pit. Their data were comparable and lower than those reported by Giordani et al. (1992). Established $\Delta Si:\Delta P$ and $\Delta Si:\Delta N$ ratios showed no consisted trend, suggesting that recycling of silicate is decoupled from the other nutrients. So far, there are no published data on nutrient fluxes for the east Adriatic.

Characteristics of the Adriatic Sea

The Adriatic Sea is the northernmost part of the Mediterranean. Its length is 783 km and average width is 243 km. According to the bathimetry (Fig. 1) the Adriatic may be divided into three parts: the shallow Northern Adriatic with an average depth of 40 m; the Central Adriatic with maximal depth of 266 m; and the Southern Adriatic basin (max. depth 1233 m). Due to the cyclonic circulation of water masses (Zore-Armanda et al., 1999), in-going streams of oligotrophic waters from the eastern Mediterranean is flowing along the east Adriatic coast and therefore, open waters of the Central and Southern Adriatic are very low in nutrient concentrations, particularly orthophosphates (Zore-Armanda et al., 1991).

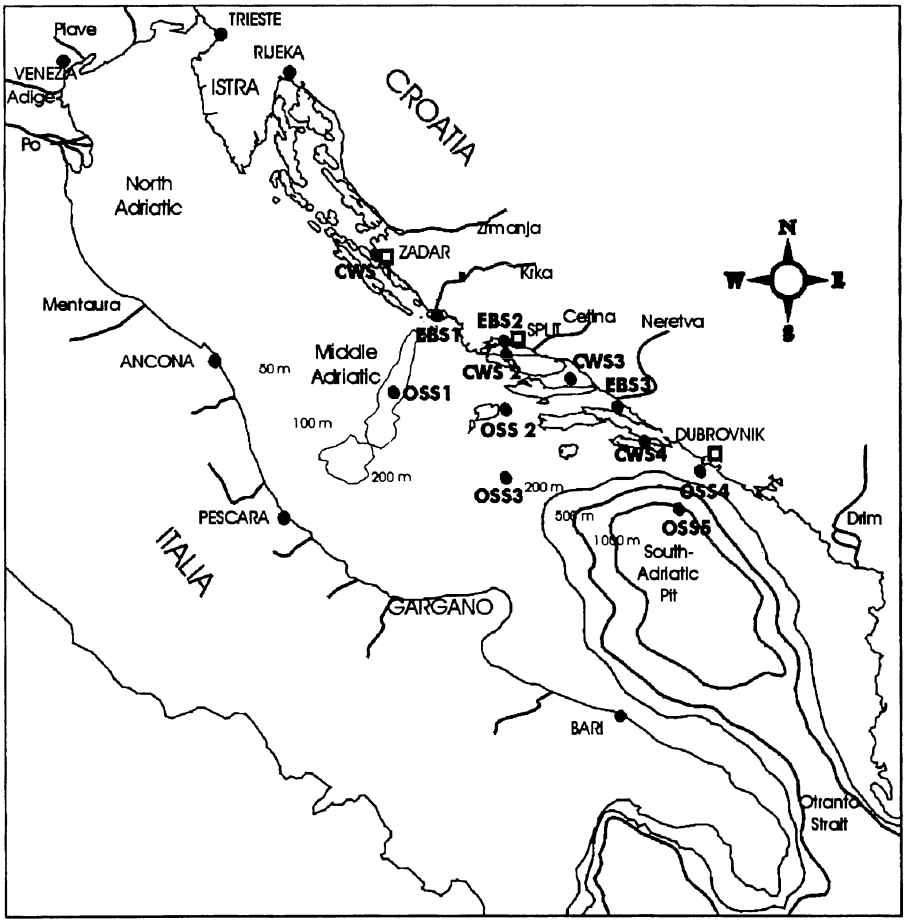

Figure 1. Map of the Adriatic Sea showing the location of sampling stations.

Numerous islands are located parallel to the eastern coast and form narrow channels which impair the exchange of water masses between the coastal area and open sea. Despite the fact that river and urban wastewater nutrient loads at the eastern Adriatic are lower compared to the load of the Po River and other Italian rivers, effects of the eutrophication process are visible in the coastal area. Since early seventies various phenomena, such as, the increase of plankton biomass (Krstulovic et al., 1997), changes in the structure of phytoplankton community (Nincevic, 2000), as well as 'red-tide' phenomena and bottom layer anoxia (Marasovic et al., 1991) occurred. Areas with the highest nutrient concentration and primary productivity are semi-enclosed bays with a high urban nitrogen and phosphorus loading (Baric et al., 1992), and salt-wedge estuaries, such as the Krka River estuary (Legovic et al., 1994), having nitrate and silicate concentration for one to two orders of magnitude higher than in open sea waters (Baric & Kuspilic, unpublished data).

This paper presents the study of benthic nutrient flux on 41 samples, collected during 1997–98 in different areas of the Central and Southern Adriatic, such as semi-enclosed bays, estuaries, channels and open waters.

Materials and methods

Sediment sampling was done between June 1997 and September 1998 at stations showed on Figure 1. A plastic gravity corer of internal diameter of 6.5 cm and length of 100 cm was used for the sampling of three parallel sediment cores for the flux study, and one more for the organic matter content and granulometry. At the same time, 10 l of seawater was sampled from the bottom layer by a plastic Nansen bottle. The seawater samples were used for the exchange of seawater above the sediment samples, as well as for the determination of dissolved oxygen in the bottom layer. CDT probe was used for the determination of temperature and salinity in the bottom layer.

Immediately after the collection, sediment samples were checked visually regarding compactness and regular stratification. Samples collected by the vertical penetration of the corer were used for the further analysis. The plastic tubes containing 40–60 cm long sediment samples and overlying seawater were closed with plastic caps and connected with Tygon tubes to a peristaltic pump and than placed in a dark thermostatic chamber with a constant temperature (bottom layer temperature). Seawater in the tubes above the sediment was first reduced to the volume of 200 ml and than slowly replaced by the flow of sampled bottom seawater at the constant rate of 100 ml min^{-1}. The replacement lasted about 30 min. Finally, the volume above the sediment samples, was adjusted to approximately 500 ml. This volume as the maximal for the accurate analytical determination of nutrient concentration changes, especially phosphate, was chosen on a series of previous experiments. The air above the seawater was removed by a continuous argon flow. The first 50 ml sample of seawater for the determination of nutrient concentration was taken 60 min after the start of incubation, consecutive ones in the intervals between 90 and 120 min, during the incubation, which lasted between 9 and 12 h. The seawater samples were analyzed on board, immediately after sampling.

Nutrient concentration was analyzed by an AutoAnalyzer II, using standard spectrophotometric methods (Strickland & Parsons, 1972; Grasshoff, 1976; Oudot & Montel, 1988) modified for this study. Wako Pure Chemical Industries (Japan) 'CSK Standard Solution' were used for the instrument calibration.

The quantity of nutrients (n) released, or uptake by sediments as a function of time (t) was calculated by the following equations (1–4):

$$n_0 = C_0 \times V_0, \tag{1}$$

$$n_1 = n_0 + (C_1 - C_0) \times V_0, \tag{2}$$

$$n_2 = n_1 + (C_2 - C_1) \times V_1, \tag{3}$$

$$n_3 = n_2 + (C_3 - C_4) \times V_2, \tag{4}$$

etc. where n_0 is initial quantity; n_1 quantity at t_1, etc. C_0 is initial concentration; C_1 concentration at t_1, etc., and V_0 is initial chamber volume; V_1 chamber volume after first subsampling, etc.

Benthic nutrient flux was calculated using the graphical method of linear regression by plotting the quantities (n; μmol) as the function of time (t; min). From the linear fitting slopes (k), the sediment surface area (A=33.166 cm^2), and a conversion factor (F=1.44$_*$10^4; μmol cm^{-2} min^{-1} to mmol m^{-2} d^{-1}) replicate fluxes (J) were established by Equation (5):

$$J = k_* A^{-1}{}_* F. \tag{5}$$

For the further calculations, an average value of the three replicate fluxes was used.

Granulometric composition of sediment samples was done according to Shepard (1954). Organic matter content (in 5 cm surface sediment samples) was determined gravimetrically after oxidation with 30% hydrogen peroxide solution and ignition at 450 °C for 6 h (Vdovic et al., 1991).

During the same sampling period, sediment samples for the determination of inorganic and total phosphorus concentration (IP, TP) were collected. The sediment samples (n = 38) were collected by gravity corer (i.d. = 3.2 cm) and the top (10 cm) of the sediment cores were divided into subsamples each 1 cm thick. The phosphorus concentrations (mmol P kg^{-1} dry sediment) were determined by the method proposed by Aspila et al. (1976).

Results

For the presentation and interpretation of obtained results, the sampling stations were grouped into three areas: estuary and bay stations (EBS), channel water stations (CWS), and open sea stations (OSS). Station depths, sediment type, organic matter content, IP and TP sediment concentrations as well as temperature, salinity and oxygen saturation in the bottom water layer are presented in Tables 1 and 2, respectively.

The open sea stations are characterised by fine-sized grain sediment, except OSS 2 where sand predominates. An increase of sand and gravel content was found at the channel water stations. At the stations in Krka estuary (EBS 1) and Neretva estuary (EBS 3), a portion of coarser sediment particles is higher than at Kastela Bay station (EBS 2). In the organic matter content, there were no distinct differences between specific areas, although high values were noticed in the each group of stations as the result of specific ecological conditions. Accordingly, EBS 1 is located in the eutrophic Krka estuary, CWS 3 in the coastal area under strong terrigenous inputs and the deepest

Table 1. Depths of stations, sediment type according to Shepard (1954), mean organic matter content ± STD, inorganic and total phosphorus concentrations ± STD through the 1997/98

Station	Depth (m)	Sediment type	Organic matter (%)	IP (mmol P kg^{-1} d. w.)	TP (mmol P kg^{-1} d. w.)
EBS 1	38	clayey silt	7.91±0.96	27.34±5.83	43.16±8.58
EBS 2	38	silty clay	5.88±0.43	13.11±3.67	28.05±5.12
EBS 3	20	clayey silt	6.25±0.38	12.99±3.35	23.33±3.90
CWS 1	38	clayey silt	5.13±0.99	5.89±1.57	13.01±3.70
CWS 2	52	clayey silt	4.91±1.34	7.75±1.59	14.17±2.32
CWS 3	66	clayey silt	8.23±1.17	n.a.[a]	n.a.[a]
CWS 4	88	clayey silt	5.41±0.93	n.a.[a]	n.a.[a]
OSS 1	206	silt	5.90±1.01	11.04±1.27	22.03±3.36
OSS 2	103	sand	2.91±0.48	13.64±4.10	19.75±2.68
OSS 3	178	clayey silt	5.63±0.39	10.51±1.97	22.51±5.67
OSS 4	100	silty clay	4.72±0.42	15.97±6.24	31.47±2.01
OSS 5	1010	silty clay	9.58±1.22	19.76±4.18	30.14±4.30

[a] Not analysed.

Table 2. Temperature (T), salinity (S) and oxygen saturation (O_2) in bottom water layer at investigated areas in cold and warm period (1997/98)

Parameter	Cold period	Warm period
T (°C)		
EBS	12.4–15.8	14.2–17.9
CWS	12.0–15.3	14.1–17.6
OSS	12.7–15.1	12.0–15.1
S (‰)		
EBS	37.98–38.40	38.21–38.27
CWS	38.01–38.45	38.01–38.39
OSS	38.49–38.65	38.48–38.82
O_2 (%)		
EBS	91.9–104.0	85.3–96.9
CWS	93.7–97.5	76.1–102.3
OSS	92.0–96.0	77.0–87.5

OSS 5 in the Southern Adriatic Pit. The only one station with the sandy sediment type is OSS 2 where the lowest organic matter content was determined. The obtained results showed very important differences in phosphorus concentrations in sediment of different stations. The lowest phosphorus sediment concentrations (both TP and IP) were found on CWS, while the highest value was at EBS 1. This extremely high TP concentration is the result of high IP content probably as the consequence of impacts from a nearby harbour terminal for phosphate ores and fertilizers.

The estimated benthic fluxes (J) at the investigated areas are shown in Table 3 for EBS, Table 4 for CWS, and Table 5 for OSS. Area flux means for the cold period of year (CP), from November to April, and the warm period (WP), from May to October, are also given.

The silicate flux was in the range from 0.16 to 2.67 mmol m^{-2} d^{-1}. The highest silicate fluxes were established at EBS 1 (river Krka estuary) which is under significant freshwater impact and known as the area with intensive diatom blooms (Legovic et al., 1996) and at EBS 2 located in eutrophic Kastela Bay (Marasovic et al., 1991; Baric et al., 1992). An increase of silicate fluxes in WP (in all investigated areas), as well as a decrease of fluxes from EBS to OSS area in the same period can be observed from the area flux means. The obtained values of phosphate fluxes were in the range between −0.031 and 0.164 mmol m^{-2} d^{-1}. The highest positive fluxes were established in EBS, particularly in WP. Negative phosphate fluxes were obtained only in CP, except once at CWS 1 (June 1997). The ammonia fluxes were between −0.51 and 2.03 mmol m^{-2} d^{-1}. Area means were mainly positive, although the negative fluxes were also obtained. The nitrate + nitrite fluxes (range: −1.32 to 1.62 mmol m^{-2} d^{-1}) showed differences between particular areas. Based on the area means, the marine sediment in OSS area is the source of ammonia and sink of nitrate+nitrite through

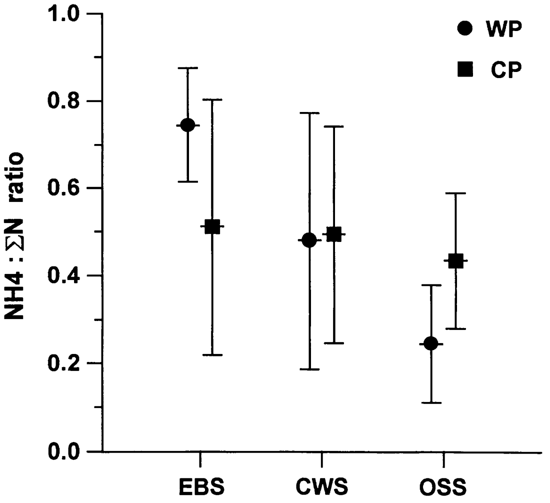

Figure 2. Ammonia:total nitrogen fluxes ratio at EBS, CWS and OSS stations in warm and cold period of the year.

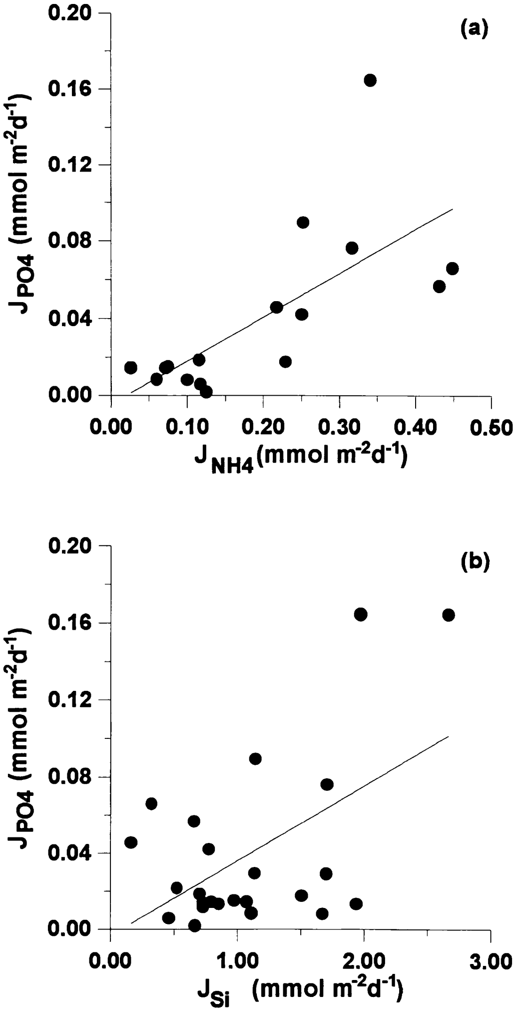

Figure 3. The relationship between phosphate and ammonia flux (a) and phosphate and silica flux (b).

the year. In CWS area, the marine sediment in CP is the source of ammonia and sink of nitrate + nitrite, while in WP the situation is opposite. In EBS area, sediment is source of ammonia while nitrate + nitrite fluxes means are zero (in CP) or positive (in WP).

Contribution of ammonia in the total benthic nitrogen exchange at the investigated areas is shown on Figure 2. In CP, the ammonia percentage in the total nitrogen exchange for all areas is 50% (remaining is nitrate + nitrite). In WP the ammonia contribution increases up to 80% in EBS and decreases to 20% in OSS.

Correlation analyses between positive nutrient fluxes (Fig. 3a, b) were statistically significant for J_{PO4} and J_{NH4}(r = 0.699, p<0.01, n =16), as well as for J_{PO4} and J_{Si} (r = 0.529, p<0.01, n = 24).

Discussion

The obtained nutrient fluxes in the Central and Southern Adriatic are comparable with and lower than worldwide published data (Hopkinson, 1987) as well as for the Northern Adriatic (Giordani et al., 1992). Low nutrient fluxes at the open sea area correspond to the low sedimentation rates (Kuptsov et al., 1981; Giordani et al., 1992; Faganeli et al., 1994).

Among all the nutrient fluxes, only silicate flux showed a regular increase from OSS to EBS area, which reflects the existing plankton density gradient in the Central Adriatic (Krstulovic et al., 1997) with a decreasing diatom portion in the phytoplankton community (68% EBS and 35% OSS; Nincevic, 2000). The silicate flux increase in WP also coincide with plankton maximas in this period (Baranovic et al., 1992; Krstulovic, 1992; Nincevic, 2000), but in shallow areas the contribution of opal solubility increase, due to higher seawater bottom temperature (Hurd, 1973) in this period of year, is also possible (Table 2). The contribution of radiolarian skeleton dissolution to the silicate fluxes in the Adriatic may be considered as negligible in areas shallower than 100 m, because these species are present in deeper areas (Krsinic, 1998). The only one unexpected high flux value (J_{Si}=1.97 mmol m^{-2} d^{-1}) was recorded on OSS 5 (March 1998). This flux increase with respect to September 1997 flux value was very probably, induced

Table 3. Benthic fluxes ± STD at EBS area

Date	Station	J_{Si} ($\mathrm{mmol\ m^{-2}\ d^{-1}}$)	J_{PO4} ($\mathrm{mmol\ m^{-2}\ d^{-1}}$)	$J_{\mathrm{NO3+NO2}}$ ($\mathrm{mmol\ m^{-2}\ d^{-1}}$)	J_{NH4} ($\mathrm{mmol\ m^{-2}\ d^{-1}}$)
17 Jun 1997	EBS 2	2.33 ± 0.14	n.a.[b]	0.28 ± 0.07	2.03 ± 1.50
20 Jun 1997	EBS 3	0.85 ± 0.22	0.013 ± 0.003	0.04 ± 0.01	−0.39 ± 0.12
27 Jun 1997	EBS 1	1.94 ± 0.25	0.013 ± 0.002	0.17 ± 0.04	−0.43 ± 0.15
16 Sep 1997	EBS 2	1.30 ± 0.11	n.a.[b]	0.18 ± 0.05	0.41 ± 0.09
25 Sep 1997	EBS 3	0.52 ± 0.32	n.a.[b]	0.09 ± 0.01	−0.12 ± 0.07
15 Dec 1997[a]	EBS 1	1.67 ± 0.71	0.008 ± 0.001	0.18 ± 0.01	0.10 ± 0.02
30 Jan 1998[a]	EBS 2	0.62 ± 0.32	−0.031 ± 0.004	−0.38 ± 0.04	0.00 ± 0.02
01 Mar 1998[a]	EBS 2	0.66 ± 0.29	0.002 ± 0.001	−0.11 ± 0.03	0.12 ± 0.09
31 Mar 1998[a]	EBS 3	0.73 ± 0.11	0.011 ± 0.011	0.14 ± 0.0	−0.20 ± 0.02
06 Apr 1998[a]	EBS 2	0.89 ± 0.38	−0.011 ± 0.007	0.05 ± 0.06	0.35 ± 0.20
09 Apr 1998[a]	EBS 1	1.14 ± 0.90	0.089 ± 0.055	0.35 ± 0.43	0.25 ± 0.24
27 Apr 1998[a]	EBS 2	1.71 ± 0.20	0.076 ± 0.043	−0.08 ± 0.05	0.32 ± 0.19
28 Jun 1998[a]	EBS 1	2.67 ± 0.24	0.164 ± 0.051	−0.16 ± 0.07	−0.36 ± 0.13
Area mean	CP	0.96 ± 0.41	0.022 ± 0.049	0 ± 0.25	0.14 ± 0.21
± STD	WP	1.60 ± 0.65	0.064 ± 0.087	0.10 ± 0.15	0.19 ± 0.95

[a] Simultaneously determined sediment phosphorus concentrations and phosphate fluxes.
[b] Not analysed.

Table 4. Benthic fluxes ± STD at CWS area

Date	Station	J_{Si} ($\mathrm{mmol\ m^{-2}\ d^{-1}}$)	J_{PO4} ($\mathrm{mmol\ m^{-2}\ d^{-1}}$)	$J_{\mathrm{NO3+NO2}}$ ($\mathrm{mmol\ m^{-2}\ d^{-1}}$)	J_{NH4} ($\mathrm{mmol\ m^{-2}\ d^{-1}}$)
21 Jun 1997	CWS 4	1.70 ± 0.06	0.025 ± 0.001	1.62 ± 0.18	−0.51 ± 0.01
26 Jun 1997	CWS 1	0.81 ± 0.25	−0.019 ± 0.009	0.03 ± 0.01	0.62 ± 0.16
24 Sep 1997	CWS 2	2.49 ± 0.41	n.a.[b]	0.09 ± 0.01	−0.09 ± 0.01
26 Sep 1997	CWS 4	1.08 ± 0.23	n.a.[b]	−0.25 ± 0.08	−0.07 ± 0.05
28 Sep 1997	CWS 3	1.37 ± 0.75	n.a.[b]	0.04 ± 0.03	−0.03 ± 0.01
14 Dec 1997[a]	CWS 1	0.46 ± 0.10	0.006 ± 0.003	0.16 ± 0.01	0.12 ± 0.02
16 Dec 1997	CWS 3	0.59 ± 0.33	−0.020 ± 0.008	−0.18 ± 0.12	0.56 ± 0.25
21 Dec 1997	CWS 2	0.70 ± 0.29	0.019 ± 0.007	0.26 ± 0.02	0.12 ± 0.03
24 Jan 1998	CWS 4	0.58 ± 0.06	−0.016 ± 0.002	0.07 ± 0.14	0.19 ± 0.13
24 Jan 1998	CWS 3	0.78 ± 0.37	0.042 ± 0.005	0.29 ± 0.05	0.25 ± 0.02
25 Feb 1998	CWS 3	0.79 ± 0.07	0.014 ± 0.002	0.20 ± 0.06	0.07 ± 0.02
26 Feb 1998	CWS 4	1.07 ± 0.52	0.014 ± 0.002	−0.15 ± 0.07	0.03 ± 0.01
28 Feb 1998	CWS 2	0.97 ± 0.03	0.015 ± 0.011	−0.28 ± 0.17	0.07 ± 0.06
30 Mar 1998	CWS 4	0.32 ± 0.04	0.066 ± 0.011	0.16 ± 0.04	0.45 ± 0.11
02 Apr 1998[a]	CWS 2	0.16 ± 0.12	0.045 ± 0.013	−0.19 ± 0.15	0.22 ± 0.09
23 Apr 1998	CWS 2	0.66 ± 0.09	0.057 ± 0.008	−0.07 ± 0.03	0.43 ± 0.26
Area mean	CP	0.66 ± 0.36	0.035 ± 0.072	−0.05 ± 0.30	0.21 ± 0.21
± STD	WP	1.49 ± 0.65	0.005 ± 0.038	0.31 ± 0.75	−0.02 ± 0.41

[a] Simultaneously determined sediment phosphorus concentrations and phosphate fluxes.
[b] Not analysed.

by the settling of particulate organic matter from the trapped cold Northern Adriatic water (Zore-Armanda, 1969) originating from one of the most productive areas in the Mediterranean (Sournia, 1973). This emphasises the higher organic matter content, average TP concentrations (Table 1) and phosphate flux (0.164

Table 5. Benthic fluxes ± STD at OSS area

Date	Station *n*	J_{Si} (mmol m^{-2} d^{-1})	J_{PO4} (mmol m^{-2} d^{-1})	$J_{\mathrm{NO3+NO2}}$ (mmol m^{-2} d^{-1})	J_{NH4} (mmol m^{-2} d^{-1})
22 Jun 1997	OSS 4	1.11 ± 0.24	0.008 ± 0.006	−0.09 ± 0.03	0.04 ± 0.06
17 Sep 1997	OSS 3	0.81 ± 0.16	n.a.[b]	0.28 ± 0.17	0.06 ± 0.03
27 Sep 1997	OSS 4	0.84 ± 0.37	n.a.[b]	−0.04 ± 0.07	−0.02 ± 0.01
27 Sep 1997	OSS 5	0.53 ± 0.40	n.a.[b]	−1.32 ± 0.74	−0.05 ± 0.02
25 Jan 1998	OSS 4	0.53 ± 0.17	0.022 ± 0.009	−0.09 ± 0.04	−0.13 ± 0.04
28 Jan 1998	OSS 3	0.73 ± 0.22	0.014 ± 0.007	0.16 ± 0.05	−0.19 ± 0.21
26 Feb 1998	OSS 4	0.52 ± 0.20	−0.012 ± 0.008	0.15 ± 0.04	−0.04 ± 0.04
28 Mar 1998	OSS 4	0.81 ± 0.04	n.a.[b]	0.24 ± 0.10	0.21 ± 0.12
28 Mar 1998[a]	OSS 5	1.97 ± 0.02	0.164 ± 0.051	−0.39 ± 0.08	0.34 ± 0.11
31 Mar 1998*a*	OSS 3	0.39 ± 0.08	−0.003 ± 0.001	−0.48 ± 0.08	0.20 ± 0.12
25 Jun 1998*a*	OSS 3	1.14 ± 0.19	0.029 ± 0.036	−0.34 ± 0.15	−0.09 ± 0.01
28 Jun 1998*a*	OSS 1	1.51 ± 0.48	0.017 ± 0.001	−0.80 ± 0.14	0.23 ± 0.06
Area mean	CP	0.83 ± 0.58	0.037 ± 0.072	−0.06 ± 0.30	0.07 ± 0.21
± STD	WP	0.99 ± 0.33	0.018 ± 0.010	−0.39 ± 0.58	0.01 ± 0.12

[a] Simultaneously determined sediment phosphorus concentrations and phosphate fluxes.
[b] Not analysed.

mmol m^{-2} d^{-1}). Since the phosphate and nitrogen (ammonia, nitrate + nitrite) flux means have not always followed plankton distribution it seems that N and P regeneration is more influenced by specific processes taking place in the surface sediment layer than by the particulate organic matter sedimentation. Probably the most interesting result of established phosphate fluxes in this part of the Adriatic is the grouping of negative fluxes in the CP. This is in the accordance to Fisher et al. (1982) who detected phosphate sorption as well as negative phosphate fluxes at lower water temperatures. We believe that the most important role in phosphate flux regulation in the Adriatic have Fe oxide-hydroxides in the oxic surface layer, representing active adsorption sites and sequestering phosphate as it was shown by Sundby et al. (1992) and Jensen et al. (1995). Since the changes in the vertical iron speciation in Adriatic sediment through the year are unknown, we assume, on the basis of oxygen bottom layer saturation ranges (Table 2), that in the CP the sediment surface layer is more oxidised than in WP. Therefore, due to a higher number of sorption sites and reduced settling of particulate organic matter on the sea floor the pore-water phosphate concentration decrease and subsequent negative P fluxes occurred at OSS 4, CWS 4 and EBS 2. The established relationships between J_{PO4} and J_{NH4} as well as between J_{PO4} and J_{Si}, in the cases of positive fluxes (Fig. 3a, b), indicate that phosphate regeneration and recovery to the water column is (after fulfilment of the sediment adsorption capacity) linear correlated to ammonia and silicate regeneration and fluxes. Similar correlation for the phosphate and ammonia flux was also found by Fisher et al. (1982) and Nixon (1981), while the correlation of silicate and phosphate was stated by Redfield, et al. (1963). The intercept of the straight lines at y axes (Fig. 3a, b) are at relative equal value (−0.0046 mmol m^{-2} d^{-1}: PO_4–NH_4 and −0.0035 mmol m^{-2} d^{-1}: PO_4-Si) and represents an average adsorption term in Adriatic sediment, while the slopes themselves could be an indicator of relative mobilities of phosphate, ammonia and silicate in sediment nutrient fluxes (Fisher et al., 1982).

The ammonia flux area means show uniform low fluxes in OSS area (maximal flux at OSS 5/ March 1998) and somewhat higher in EBS area. The relationship between J_{PO4} and J_{NH4} (Fig. 3a) with an ΔJ_{PO4}:ΔJ_{Si} ratio of 1:4.4 which is significant lower to the Redfield ratio (Redfield et al., 1963) points to significant removal of ammonia from the pore-water by adsorption (Rosenfeld, 1979) or nitrification. The main characteristic of ammonia fluxes in the Adriatic is occurrence of negative fluxes. The occurrence of negative ammonia fluxes has been reported by other authors (Simon, 1988; Rowe & Phoel, 1992; Lohse et al., 1995). However, an explanation for this phenomenon was only provided by Simon (1988). He related negative fluxes to the sediment resuspension. An occasional ammonia sediment uptake was proposed by Laima (1992) studying seasonal changes of ammonia pore water profiles and sediment adsorption capacity for ammonia in the coastal area of Denmark. He determined a high sediment adsorption capacity (adsorption K-value up to 108), but the relation between sediment ammonia uptake and sediment characteristics was not found. Our obtained negative ammonia fluxes are probably the result of different factors specific for the each studied area. In OSS area, with low primary production rate ($\sim$280 mg C m^{-2} d^{-1}) (Miseta, 1999) ammonia is probably completely oxidised into nitrite and nitrate at the oxic surface sediment layer. In the shallow channel area, the negative ammonia flux is related to the higher silicate flux (Table 3). If we use the increased silicate flux as the indicator of increased organic matter settlement, than the ammonia adsorption onto detritic organic matter would be the main controlling process in the direction of ammonia fluxes either into or out of the sediment, as proposed by Rosenfeld (1979). In addition, the ammonia uptake is probably stimulated by the relative high bottom current (indicated by prevailing coarsesized sediment particles) causing the sediment resuspension, as proposed by Simon (1988). In the estuary and bay areas (Table 4) the negative fluxes were obtained at stations EBS 1 and EBS 3 but not at station EBS 2. The difference may be related to the rivers impact. Krka River (EBS 1) and Neretva River (EBS 3) are much larger rivers than Jadro River (EBS 2) and granulometric sediment analysis at EBS 1 and EBS 3 showed higher portion of coarsesized particles (Table 1) which indicates the existence of significant counter-current in the bottom layer and consequent sediment resuspension.

Similar to the ammonia, nitrate + nitrite showed also negative fluxes, especially in the OSS area in WP (area with lowest oxygen saturation ranges; Table 2). Such negative fluxes are the indicators of intensive denitrification, which occur after the reduction of oxygen and Mn (IV) (Froelich et al., 1979), but as shown by Brandes & Devol (1995) denitrification can take place even in the presence of oxygen.

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Nitrogen fluxes and budget seasonality in the Ria Vigo (NW Iberian Peninsula)

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Abstract

Inorganic and organic nitrogen fluxes in the Ria Vigo have been quantified in order to recognise the contrasting nitrogen budget scenarios and understand the biogeochemical response to eutrophication events. According to the nitrogen biogeochemical pathways of the ria reservoir (photosynthesis, remineralization, denitrification, PON rain rate and sedimentation), three main seasonal behavioural trends are emphasised: (1) low inorganic nitrogen inputs and low organic nitrogen fluxes, (2) high inorganic nitrogen input and output, (3) high inorganic nitrogen input and high organic nitrogen output. The first scenario occurs in late spring and in summer during non-upwelling situations. The consumption of inorganic nitrogen by net photosynthesis is approximately 2 mol N s^{-1} and the ria is oligotrophic (12 mgC m^{-2} h^{-1}). The outgoing estuarine residual current transports phytoplanktonic material towards the mouth of the ria whereupon it sediments and is remineralized as it falls to the lower water layers and the incoming residual current. The regenerated nitrogen is reintroduced to the photic ria layer which leads to the greatest reduction in dissolved oxygen concentration (50% of saturation). Recycled nutrients play an important role in primary production during this oligotrophic state of the ria. Thus, approximately half of the inorganic nitrogen utilised by photosynthesis is ammonium. The majority of PON is deposited inside the ria (0.8 mmol N m^{-2} d^{-1}) and the denitrification rate is 0.3 mmol N_2 m^{-2} d^{-1}. The other two cases occur in winter and spring–summer with upwelling. In winter, estuarine circulation and freshwater contributions control the nitrogen cycle. The ria mainly exports nitrate (up to 14 mol N s^{-1}) and so there is fertilisation but no eutrophication. In spring and summer, the nitrogen cycle is controlled by upwelling circulation. The inorganic nitrogen consumption by net photosynthesis is high, 7–14 mmol N m^{-2} d^{-1}, and the ria is a natural eutrophic system (70 mgC m^{-2} h^{-1}). Accordingly, 90% of organic nitrogen is synthesised from nitrate and the upwelling-increased circulation exports 6.5 mol N s^{-1} of organic nitrogen.

Introduction

Temperate estuaries situated on eastern oceanic coastlines, such as the Ria Vigo in northwest of Iberian Peninsula, are normally characterised by substantial rates of marine production. Nutrient contamination in the northwestern Spanish estuaries, also known as the Galician rias, is relatively low (Prego et al., 1999) and therefore, mainly due to summer upwelling, the oceanic influence inside the rias may be important (Prego & Bao, 1997). On the contrary, in open western marginal estuaries, for instance Chesapeake Bay (Nixon, 1987), large rivers, agricultural, anthropogenic, and atmospheric influences are the main reason for the high nutrient inputs. However, fertilisation of rias arises from upwelling when ria residual velocities may be tripled (Prego & Fraga, 1992). In this way, the river-ria-ocean system could offer useful information in relation to natural fertilisation processes (Prego, 1994) and act as a good benchmark for hyper-eutrophied systems.

In the Ria Vigo, the average annual primary production is 350 mgC m^{-2} d^{-1} (Vives & Fraga, 1961) within which nitrogen plays the usual key role in seawater biological processes (McCarty & Carpenter, 1983; Paasche, 1988). In rias, because the variation in phosphate does not correlate with variations in biomass (Fraga, 1960), inorganic nitrogen is the most

Table 1. Nitrogen (μmol N kg^{-1}), oxygen (μmol O_2 kg^{-1}) and NO (μmol O_2 kg^{-1}) concentrations in the Ria Vigo reservoir

		31-I	28-II	5-III	26-V	31-V	7-VII	12-VIII	4-IX	21-IX	3-X
Inorganic nitrogen											
Nitrate											
Incoming	C_i	4.79	5.05	4.17	5.76	8.79	5.96	3.83	10.73	1.42	7.85
Outgoing	C_o	5.71	10.94	5.73	0.50	3.34	0.22	0.19	1.99	0.53	0.73
Descending	C_d	6.32	11.17	5.74	0.35	3.80	0.25	0.50	2.54	1.25	3.42
Ascending	C_a	4.66	6.91	4.48	3.31	8.23	1.68	0.86	7.76	1.51	5.14
Freshwater	C_w	28.3	26.8	23.8	17.8	22.1	20.2	40.1	20.0	10.8	16.7
Nitrite											
Incoming	C_i	0.63	0.36	0.61	0.39	0.46	0.22	1.12	0.20	0.18	0.67
Outgoing	C_o	1.01	0.26	0.37	0.10	0.39	0.01	0.09	0.49	0.16	0.13
Descending	C_d	1.04	0.27	0.37	0.07	0.33	0.02	0.18	0.25	0.25	0.50
Ascending	C_a	1.00	0.33	0.51	0.28	0.44	0.10	0.46	0.50	0.28	0.71
Freshwater	C_w	0.6	0.9	0.8	1.1	2.0	0.0	0.0	0.0	0.8	0.0
Ammonium											
Incoming	C_i	0.98	1.56	0.33	1.04	0.18	4.11	2.10	1.71	2.67	1.79
Outgoing	C_o	3.81	2.55	0.78	0.21	0.15	1.69	2.00	0.91	0.39	0.45
Descending	C_d	3.45	3.37	0.97	0.47	0.15	0.24	1.46	0.81	2.44	3.61
Ascending	Ca	1.38	2.55	0.89	1.93	0.23	1.51	1.59	1.78	3.84	4.55
Freshwater	C_w	8.9	7.3	14.3	30.1	72.5	131.0	148.0	135.5	22.2	93.5
Organic nitrogen											
Incoming	C_i	8.69	7.48	4.34	5.52	5.72	5.00	11.96	6.14	9.03	4.59
Outgoing	C_o	11.28	11.78	5.72	8.74	7.53	8.53	15.95	13.19	12.53	13.41
Descending	C_d	10.12	10.42	5.49	10.44	6.97	9.91	13.04	15.46	12.53	9.72
Ascending	C_a	8.64	7.66	4.27	7.89	4.78	9.66	13.78	8.76	9.93	7.07
Freshwater	C_w	10.6	7.3	11.6	26.7	40.0	95.0	180.0	95.0	28.5	90.0
Oxygen											
Incoming	C_i	219.0	244.7	248.1	215.6	205.2	169.1	180.4	177.7	217.3	160.5
Descending	C_d	254.0	260.3	243.1	272.9	243.6	246.0	200.1	255.4	227.0	195.0
Ascending	C_a	223.5	239.4	244.0	222.7	206.1	215.6	195.9	196.2	206.8	162.8
NO											
Incoming	C_i	283	313	300	287	302	277	248	304	266	263
Descending	C_d	358	406	314	281	287	257	219	291	263	265
Ascending	C_a	293	335	302	276	297	246	206	296	257	260

adequate nutrient for considering the seasonal changes of photosynthesis, remineralization and sedimentation processes.

Nutrient salt concentrations in the Galician rias vary seasonally (Prego et al., 1999) as qualitatively described in coastal areas with temperate climates (Vaccaro, 1965). Furthermore, the ria nitrogen cycle is similar to the one given by Aston (1980) for an estuary. Therefore, it is necessary to quantify the nitrogen cycle by surveying possible relations to seasonal changes and hydrographical conditions throughout the year, that is the objective of this paper.

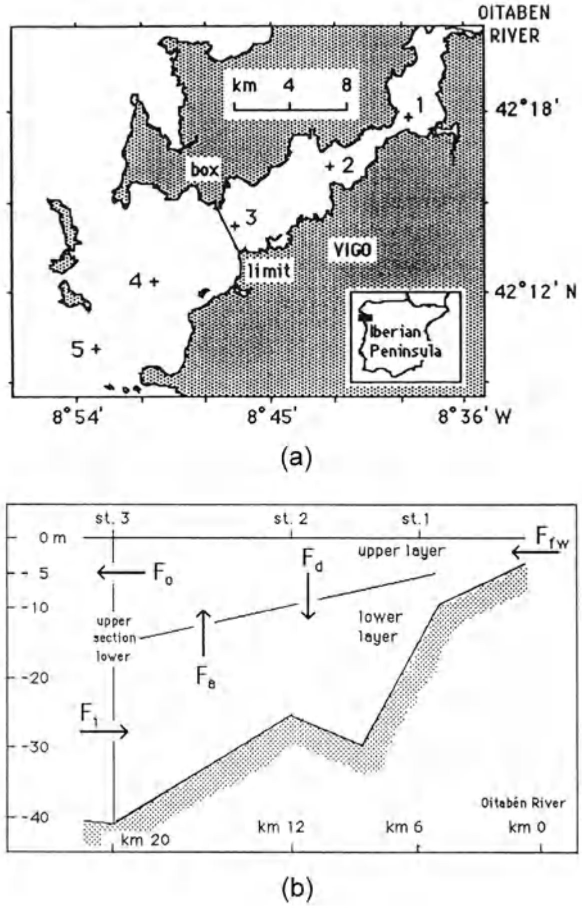

Figure 1. The Ria Vigo and its budget model. Ria-ocean exchange fluxes are the incoming from subsurface (F_i), and the outgoing from surface (F_o); layer interchange fluxes are ascending (F_a) and descending (F_d); and there are also freshwater contributions (F_w).

Survey area and data

The Ria Vigo (Fig. 1) is the southern-most ria in Galicia (NW of the Iberian Peninsula) and similar to the other three Rias Bajas. The main fluvial contribution is from the Oitabén river at it's head, making up 75% of the continental water contributions to the ria. The mean annual flow is 20 m^3 s^{-1}, varies from 55 m^3 s^{-1} in February to 2 m^3 s^{-1} in August.

The ria is a partially stratified estuary where depth and salinity increase seawards. However, the external part of the ria is under oceanic influence (Prego & Fraga, 1992) and in this paper the inner and middle ria parts will be considered as the biogeochemical nitrogen reservoir (stations 1–3, Fig. 1), of length 20 km, area 56 km^2 and volume 0.8 km^3.

Sampling in the Ria Vigo was carried out at five stations along the ria axis (Fig. 1). Ten transects were made from January to October 1986. Salinity, temperature, nitrate, nitrite, ammonium, dissolved organic nitrogen (DON), oxygen and chlorophyll *a* were measured. These data have been compiled and published previously by Prego et al. (1988).

Fluxes, biogeochemical budget and results

The present study of nitrogen biogeochemical cycling in the Ria Vigo (Fig. 1) is based on the general budgeting procedure applied to non-conservative materials (Gordon et al., 1996):

$$d[N]/dt = \Sigma F + \Delta N, \qquad (1)$$

where d[N]/d*t* is the time variation in the mass of nitrogen present in the reservoir; ΣF is the sum of the inputs (positive) and outputs (negative) of the nitrogen fluxes in the water flows (advection and mixing) to and from the reservoir; and ΔN are the non-conservative processes acting within the reservoir which add and/or remove nitrogen, i.e. biogeochemical processes. These non-conservative processes could be quantified with a steady state assumption of the ria nitrogen cycle, from which the derivative d[N]/d*t* is equal to zero.

Water exchange between the Ria Vigo and the offshore coastal zone is driven by positive residual circulation. Thus, seawater entering the ria near the bottom (lower layer) mixes with outgoing fluvial surface water and then exits the ria in the upper layer. Accordingly, a two layer reservoir model for quantifying this transport of water in the ria has been proposed for the Ria Vigo by Prego & Fraga (1992) based on the contribution of fresh water and salinity. The flows of incoming and outgoing, ascending and descending water in this ria have been calculated (Table 2).

The main concentrations of inorganic (nitrate, nitrite, ammonium) and organic nitrogen in the above water flows were calculated from ria data (Prego et al., 1988) and then area-averaged in the case of the ria-ocean separation section (i.e. C_i and C_o, Table 1) or volume-averaged for each two layer section (i.e. C_a and C_d, Table 1). Freshwater nitrogen concentrations (C_w) data (Prego, 1994) were calculated from non-polluted river water and polluted water from Vigo sewage.

The resulting fluxes for the ria are summarised in Table 2 and correspond (Fig. 1) to the fluxes of inorganic and organic nitrogen in freshwater (F_w) and

Table 2. Residual water flows (Q in 10^3 kg s^{-1}) and nitrogen (mol N s^{-1}) and NO (mol O_2 s^{-1}) fluxes in the Ria Vigo reservoir

		31-I	28-II	5-III	26-V	31-V	7-VII	12-VIII	4-IX	21-IX	3-X
Water											
Incoming	Q_i	1045	850	861	174	699	182	214	484	862	246
Outgoing	Q_o	1098	989	898	183	704	184	215	486	874	249
Descending	Q_d	161	263	193	69	122	234	595	1012	651	359
Ascending	Q_a	1206	1113	1054	242	829	416	809	1496	1513	605
Freshwater	Q_w	53	139	37	9	5	2	1	2	13	3
Inorganic nitrogen											
Incoming	F_i	6.68	5.94	4.40	1.25	6.58	1.88	1.51	6.27	3.67	2.54
Outgoing	F_o	11.57	13.75	6.17	0.15	2.74	0.53	0.49	1.51	0.94	0.33
Descending	F_d	1.76	3.90	1.37	0.06	0.52	0.13	1.28	3.64	2.57	2.71
Ascending	F_a	8.48	10.91	6.20	1.34	7.30	1.37	2.36	15.03	8.52	6.29
Freshwater	F_w	2.00	4.86	1.44	0.44	0.48	0.30	0.29	0.31	0.44	0.33
Organic nitrogen											
Incoming	F_i	9.08	6.36	3.74	0.96	4.00	0.91	2.56	2.97	7.78	1.13
Outgoing	F_o	12.39	11.65	5.14	1.60	5.30	1.57	3.43	6.41	10.95	3.34
Descending	F_d	1.63	2.74	1.06	0.72	0.85	2.32	7.76	15.65	8.16	3.49
Ascending	F_a	10.42	8.53	4.50	1.91	3.96	4.02	11.15	13.11	15.02	4.28
Freshwater	F_w	0.56	1.01	0.43	0.24	0.20	0.19	0.18	0.19	0.37	0.27
NO											
Incoming	F_i	295	266	258	50	211	49	53	147	221	65
Outgoing	F_d	58	107	61	19	35	59	130	294	171	95
Descending	F_a	353	373	319	67	246	103	180	442	389	158

marine water (F_i), outflux (F_o), ascending (F_a) and descending flux (F_d) between the layers.

Equation (1) may be applied to the Ria reservoir budget:

(a) To the fluxes (ΣF):

Inorganic nitrogen, upper layer :

$$\Sigma F_{iu} = F_w - F_o + F_a - F_d$$

Inorganic nitrogen, lower layer :

$$\Sigma F_{il} = F_i - F_a + F_d$$

Organic nitrogen, upper layer :

$$\Sigma F_{ou} = F_w - F_o + F_a - F_d$$

Organic nitrogen, lower layer :

$$\Sigma F_{ol} = F_i - F_a + F_d$$

(b) To the biogeochemical processes (ΔN):

Inorganic nitrogen, upper layer :

$$\Delta N_{iu} = P$$

Inorganic nitrogen, lower layer :

$$\Delta N_{il} = R$$

Organic nitrogen, upper layer :

$$\Delta N_{ou} = -P + D$$

Orgenic nitrogen, lower layer :

$$\Delta N_{ol} = -R - D + S + T$$

Only approximately 1% of light penetrates to the lower layer. Therefore, photosynthesis occurs mainly in the upper layer and remineralization occurs in the lower layer. 'P' is 'net production' according to Cooper's definition (Cooper, 1933) since 'P' is the result of photosynthesis plus respiration in the upper layer. 'P' must be negative as PON is photosynthesised at the expense of inorganic nitrogen. 'R' is the net remineralization in the lower layer. It must be positive since inorganic nitrogen appears from PON in this layer. In the budget, the exchange of PON between layers is established by '+D' in the third (b) equation and, clearly, the opposite '−D' in the fourth equation. Hence, 'D' will be negative if PON falls to the lower

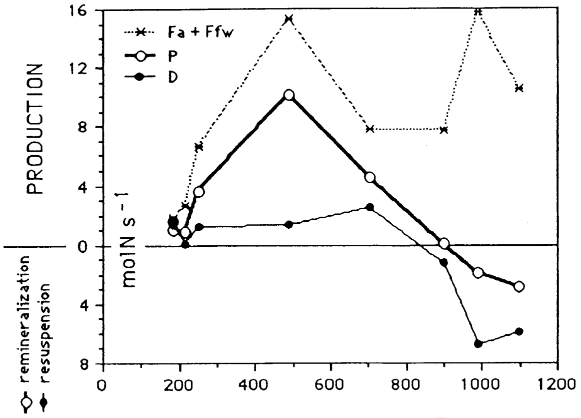

Figure 2. Values of primary production (P as net production, D as new production and $F_a + F_w$ as inorganic nitrogen inputs) *versus* the outgoing water flow (Q_o) in the Ria Vigo reservoir.

layer and must be the 'new production', according to Eppley (1989), which is measured by PON collected in sediment traps at the base of the photic zone. Net sedimentation in the lower layer is '+S'. This value will be negative if PON leaves this layer, and positive if there is sediment resuspension.

There are five unknowns, P, R, D, S and T (denitrification) in (b) and four equations in (a). Therefore, one biogeochemical flux must be known in order to solve the system. This will be denitrification, and may be resolved if the 'NO' parameter is considered.

The compositional relation for O:C:N:P of 175:129:17:1 has been reported for the phytoplankton of the Ria Vigo (Ríos & Fraga, 1987). From this ratio and the various nutrient salts and inorganic carbon in seawater, the following equation is derived:

$$\begin{aligned} &129\,\mathrm{HCO_3^-} + 17\,\mathrm{NO_3^-} + \mathrm{HPO_4^{2-}} + 126\,\mathrm{H_2O} \\ &= (\mathrm{C_{129}H_{234}N_{17}O_{70}P}) + 175\,\mathrm{O_2} + 148\,\mathrm{OH^-}. \end{aligned} \tag{2}$$

When selecting a component of each part of the photosynthesis-remineralization equation, i.e. O_2 and NO_3^-, and its stoichiometric quotients, 175:17 (10.29), the resulting parameter, 'NO', (Broecker, 1974) is unaffected by photosynthesis and remineralization processes, but it is for denitrification. Thus:

$$\begin{aligned} \text{'NO'} &= [\mathrm{O_2}] + 10.29[\mathrm{NO_3^-}] \\ &\quad + 9.79[\mathrm{NO_2^-}] + 8.29[\mathrm{NH_4^+}], \end{aligned} \tag{3}$$

where the correction terms for nitrite and ammonia are a recognition that these two compounds can reduce the calculated oxygen production in Equation (2) during photosynthesis or remineralization.

Now, the 'NO' budget in the lower layer will give the denitrification values:

(c) 'NO', lower layer:

$$\begin{aligned} \Sigma F_{ol} &= F_i - F_a + F_d, \\ \Delta N_{ol} &= T, \end{aligned}$$

where the NO fluxes (Table 2), as previously mentioned, are the water flows (Table 2) multiplied by the NO concentrations (Table 1).

Finally, the budget and the biogeochemical processes (P, R, D, S and T) can be quantified (Table 3).

Discussion

To describe the seasonal trends in the nitrogen budget in the Ria Vigo, three typical scenarios are observed according to the nitrogen ocean-ria-river exchanges obtained in Tables 2 and 3 and the nutrient salt trends in Galician rias (Prego et al., 1999).

Late spring and summer without upwelling

In the Ria Vigo reservoir from May to September when residual flow is low ($Q_o < 300 \cdot 10^3$ kg s^{-1}, Table 2), the inorganic nitrogen input to the photic layer ($F_a + F_w$, Table 2) is between 1 and 3 mol N s^{-1}. Nitrogen consumption by net photosynthesis (P in Table 3) and PON fall (D in Table 3) have similar values and these three fluxes are closed in Fig. 2. In other words, the low input of inorganic nitrogen is almost totally utilised for organic nitrogen synthesis by phytoplankton and PON sinks in the same area where it is photosynthesised. Since the interior ria is 56 km^2 and C:N is 129:17 as Equation (1), the nitrogen participation in primary production is equivalent to 140 mgC m^{-2} d^{-1}, i.e. approximate to open water production. This ria behaviour is similar to a mesotrophic system near to oligotrophy (Dugdale & Wilkerson, 1992) where recycled nutrients play an important role in primary production (Dugdale & Goering, 1967). Thus, the ammonium budget (from Table 2 data) in the photic layer suggests that half of inorganic nitrogen used by photosynthesis is ammonium, a similar percentage to the open ocean (Gilbert, 1988).

In accordance with the chlorophyll distribution (Fig. 3b), phytoplanktonic material is present in the outgoing (upper 10 m) and in the incoming ria flow.

Table 3. Nitrogen biogeochemical processes (mol N s^{-1}) in the Ria Vigo reservoir. A negative value of P indicates photosynthesis of PON since inorganic nitrogen is used and disappears, and a positive value of P indicates remineralisation; the opposite occurs with remineralisation. A negative value of D and S indicate transfer of organic nitrogen from the upper to the lower layer, or from the lower to the sediment, respectively. Negative values of T are denitrification (units in mol N_2 s^{-1}) since nitrogen exits the lower layer

		31-I	28-II	5-III	26-V	31-V	7-VII	12-VIII	4-IX	21-IX	3-X
Upper layer											
Photosynthesis	P	+2.9	+1.9	−0.1	−1.6	−4.5	−1.0	−0.9	−10.2	−5.5	−3.6
Fall of PON	D	+ 5.9	+6.7	+1.2	−1.4	−2.5	−1.3	−1.0	−1.4	−1.7	−1.3
Lower layer											
Remineralisation	R	0.0	+1.1	+0.4	+0.1	+0.2	−0.6	−0.4	+5.1	+2.3	+1.0
Sedimentation	S	+5.6	+7.2	+1.3	−0.8	−3.2	−0.7	−0.2	−1.8	−0.1	−0.4
Denitrification	T	0.0	0.0	0.0	−0.3	0.0	−0.5	−0.4	0.0	−0.3	−0.2

Error is estimated at ±0.1 mol N s^{-1} and 1 mol N s^{-1}=21.6 mgN m^{-2} d^{-1}.

The 1% light penetration depth (22 m) allows inorganic nitrogen consumption in the lower layer and photosynthesis is indicated by negative values, i.e. net remineralization occurs: −0.6 and −0.4 mol N s^{-1} at July 7th and August 12th, respectively (Table 3).

This ria behaviour in late spring and summer without upwelling could be interpreted as phytoplankton transport in the outgoing current towards the mouth of the ria whereupon it falls to the lower layer and is re-introduced to the upper layer at the head of the ria by the incoming current. Consequently, a substantial part of phytoplankton and particulate matter traces an elliptical trajectory and is maintained within this area of the ria (Fig. 3b). Contemporaneously, the biomass probably varies slightly, and PON must fall to the sediment (0.8 mmol N m^{-2} d^{-1}) mainly in this zone. This description is illustrated in Figure 4c where the freshwater nitrogen contributions, pristine plus antropogenic, are very low compared with ocean inputs.

A non-zero NO balance is observed in the lower layer. There is denitrification in the sediment with N_2 production at around 0.3 mol s^{-1} (Table 3). In accordance with the ria dimensions, this value corresponds to 0.5 mmol N m^{-2} d^{-1}, a similar flux to reported estuarine values using different methods (Koike & Sorensen, 1988). This situation does not lead to ria anoxia. In accordance with Figure 5, oxygen saturation shows a linear relationship with DON concentration:

$$\% \, O_2 = 49 + 3.73[\mathrm{DON}] \quad r = 0.85,$$

where, in the hypothetical case of total depletion of DON, only the 50% oxygen saturation would remain. Nevertheless, as the lowest DON concentration measured is the water column is greater than 4 μm (Prego et al., 1988), the usual values of oxygen saturation could be higher than 65% in the ria during summer non-upwelling conditions. Therefore, denitrification is important in the ria, as in July (Table 3), and anoxia is not attained under natural conditions.

Winter

A high supply of inorganic nitrogen occurs during the rainy season (Fig. 3a) when residual flow in the ria is over 800 · 10^3 kg s^{-1}. During high run off events, the bed sediments are resuspended to the water column and, only in winter (S in Table 3), sedimentation has a positive sign: 8.6 mmol N m^{-2} d^{-1}. Sediment eventually reaches the upper layer of the ria since fall is also positive (D in Table 3) and remineralization occurs throughout the water column (positive P and R, Table 3). During this typical winter behaviour (illustrated in Fig. 4a for February 28th) the inorganic nitrogen input from river and sediments is relatively high, i.e. the fertilisation effect is important. However, nitrogen leaves the ria as exports of inorganic and organic nitrogen, 12–14 and 11–12 mol N s^{-1}, respectively. The Ria Vigo is flushed of eutrophic nutrients by water advection as occurs in estuaries during periods of high fluvial flow (Meybeck & Helmer, 1989).

Spring and summer with upwelling

Another scenario of high supply of inorganic nitrogen in the ria occurs under upwelling conditions in either spring or summer. In accordance with Prego & Fraga (1992) this is exemplified on May 31st and September 4th cruises. Circulation in the ria is in the intermediate flow range (from 300 · 10^3 to 800 · 10^3 kg^{-1}, Fig.

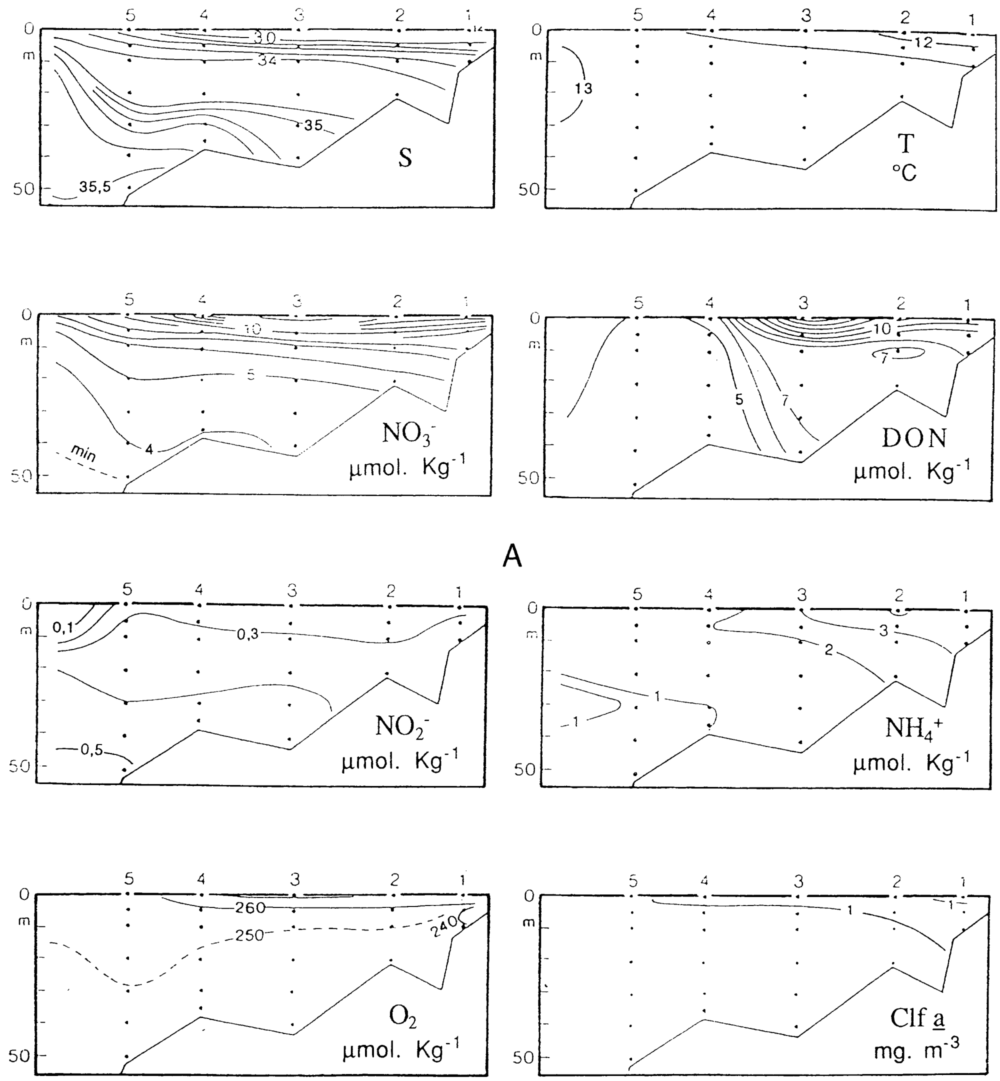

Figure 3. Isopleth maps of salinity, temperature, nitrate, nitrite, ammonium, dissolved organic nitrogen, oxygen and chlorophyll *a* during three typical scenarios in the Ria Vigo. (A) winter, 28th February; (B) summer with relaxed upwelling, 12th August; (C) summer with upwelling, 4th September.

2). Upwelling produces a high nitrate ria input (7–15 molN s^{-1}, Table 2 and Fig. 2). Nitrate in the ria is not totally used (Fig. 4b) by phytoplankton. Therefore, nitrogen consumption in net photosynthesis is less than the inorganic nitrogen inputs (Fig. 2) and there is exportation of nitrogen to offshore waters (2 mol N s^{-1}, Table 2). Photosynthesis uses 7–14 mmol N m^{-2} d^{-1} (1000 mgC m^{-2} d^{-1}) i.e. 10 times greater than during the mesotrophic case, and the ria becomes eutrophic. However, eutrophication is a natural process in the ria and is typical for upwelling areas (Dugdale & Wilkerson, 1992). The low ammonium budget (from Table 2 data) in the photic layer shows the trivial importance of recycled nutrient (9–16% DIN) in net photosynthesis. Phytoplankton consumes 65% of the inorganic nitrogen entering the upper layer of the interior part of the ria, as occurs in the upper estuary of Rhode River, U.S.A. (Jordan et al., 1991). This high production does not cause a greater sinking of PON (14%) in summer, but remains quantitatively similar to the data in July or August (Fig. 2) since there is high exportation of organic nitrogen (6 mol N s^{-1}, Table 2). Outside the ria PON is remineralized and reintroduced in the ria by the incoming upwelling current (Prego, 1994).

The increased circulation in the ria due upwelling (Prego & Fraga, 1992) is the main reason for PON

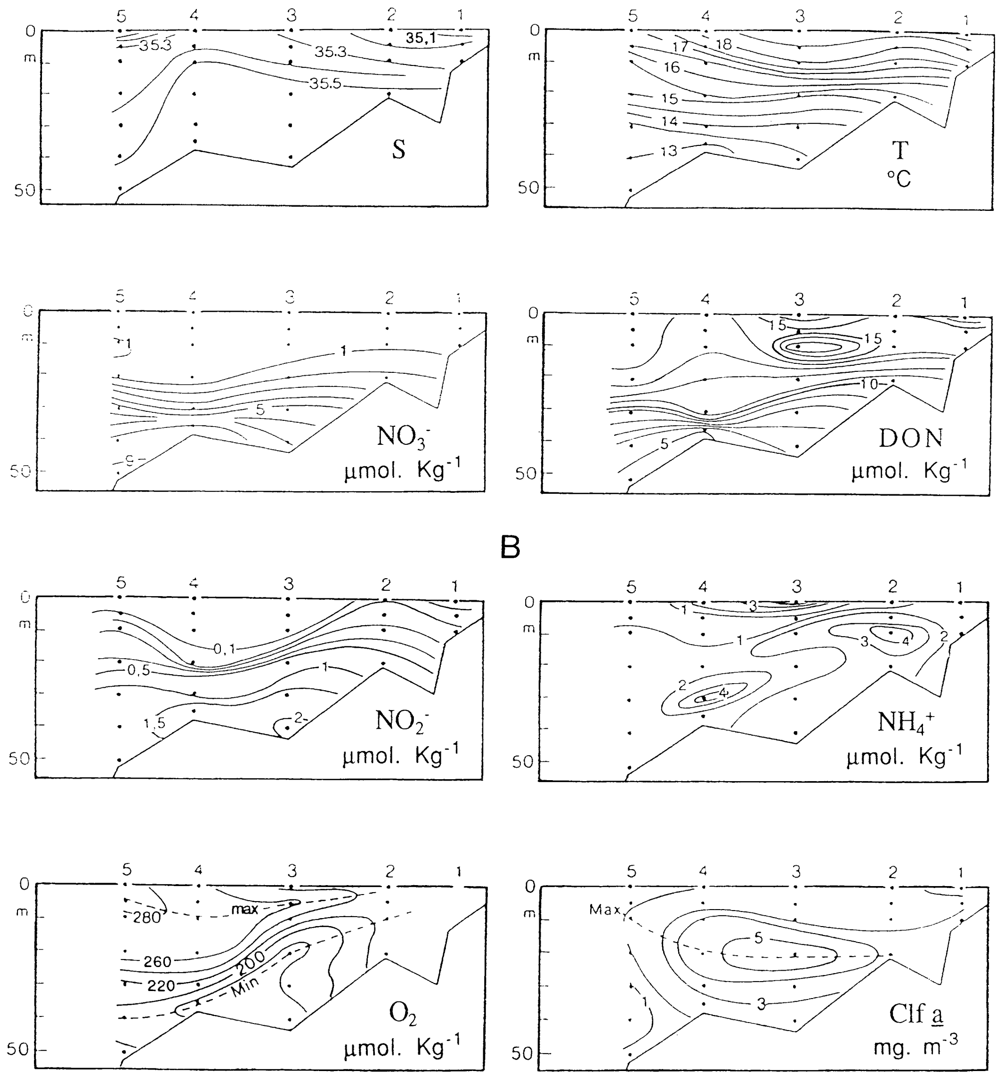

Figure. 3B

export in this situation. The elliptic cell during low residual circulation, non-upwelling summer states, is disrupted leading to an open flux of organic nitrogen to the outer ria, Figures 3c & 4b.

Conclusions

The Ria Vigo shows similar seasonal trends of nitrogen cycling as a temperate estuary. Three different scenarios are observed. In winter, the residual circulation is high and the ria mainly exports up to 14 mol s^{-1} nitrate. In spring, there is a phytoplankton bloom and net photosynthesis consumes inorganic nitrogen in the region of 2–4 mol N s^{-1}, and therefore the ria exports little DIN (<0.5 mol N s^{-1}). In late spring or summer with non-upwelling, ammonium accounts for half of the DIN used in photosynthesis. Net production of PON is low (1 mol N s^{-1}) and the majority of organic material sediments inside the ria where benthic processes drive denitrification at 0.5 mmol N m^{-2} d^{-1}.

The Ria Vigo, as other estuaries on the eastern Atlantic coast under seasonal upwelling events, displays another behaviour caused by upwelling inside the ria. Upwelling results in high net photosynthesis of organic nitrogen (10 mol N s^{-1}) which is mainly exported from the ria at up to 6.5 mol N s^{-1}, or sedimented at 50 mg N m^{-2} d^{-1}.

In agreement with these inorganic nitrogen contributions, the Ria Vigo could be considered

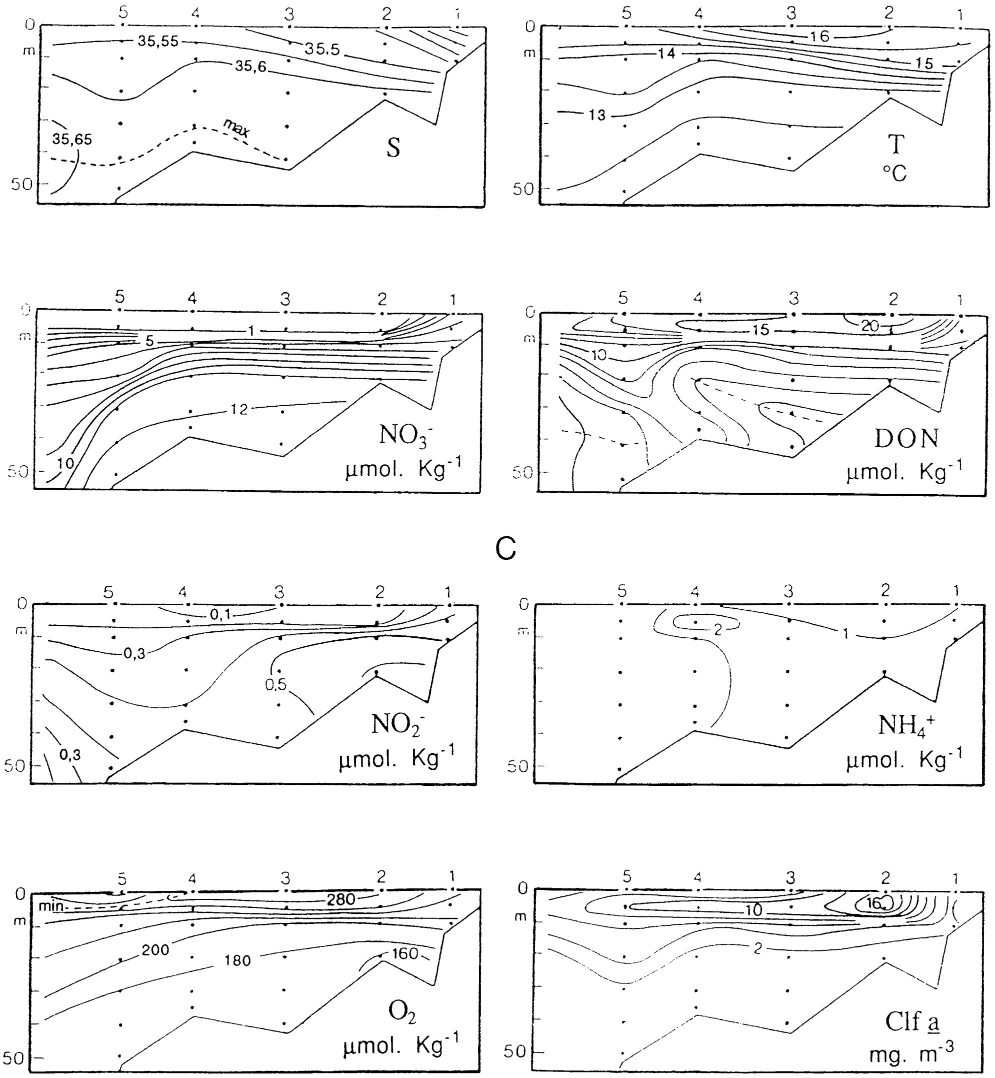

Figure. 3C

oligotrophic-eutrophic, mainly depending on upwelling. Whenever the contribution of inorganic nitrogen to the Ria Vigo is high there is always a high nitrogen output. The ria acts like a pristine coastal system because anthropogenic and fluvial inputs are low in comparison with the eutrophic events caused by seasonal upwelling. In this way, no natural anoxic episodes occur in the rias and the river-ria-ocean system offers useful information in relation to natural fertilisation processes and serves as a good referenced for hyper-eutrophied systems.

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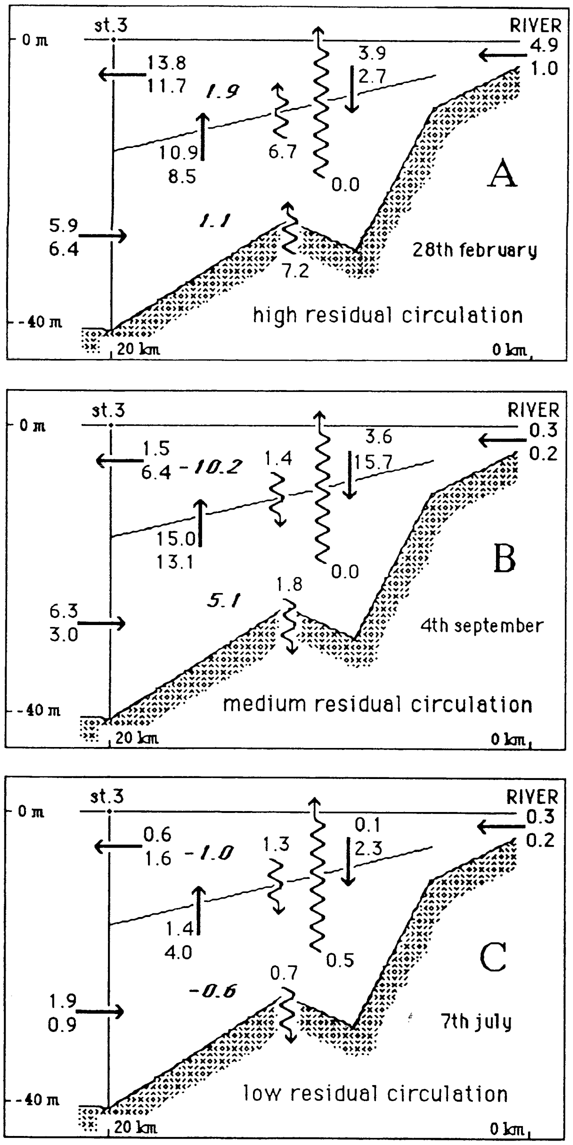

Figure 4. Nitrogen fluxes and budget in three normal ria situations according to the residual flows. (A) high DIN input and output (winter); (B) high DIN input and DON output (upwelling); (C) low DIN input and DON output (spring and summer without upwelling). Units: mol N s^{-1}. Normal arrows: upper numbers are DIN and lower DON fluxes. Wavy arrows: PON fall, sedimentation and denitrification. Italic numbers: photosynthesis (negative values) and remineralization (positive values).

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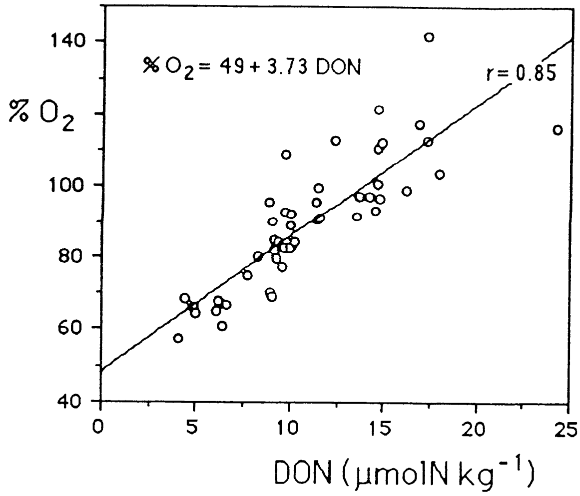

Figure 5. Saturation percentage of dissolved oxygen *versus* dissolved organic nitrogen concentration in the Ria Vigo during summer.

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The winter and summer partitioning of dissolved nitrogen and phosphorus. Observations across the Irish Sea during 1997 and 1998

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Key words: Irish Sea, total oxidised nitrogen, soluble reactive phosphate, dissolved organic nitrogen, dissolved organic phosphorus, N:P ratios

Abstract

Winter and summer water column profiles from a transect co-inciding with the 54° N parallel across the Irish Sea were examined for relationships between the fractions of total dissolved nitrogen and phosphorus. Inorganic nitrogen and phosphorus gave the expected seasonal variation. As inorganic nitrogen concentrations decreased during the summer, dissolved organic nitrogen concentrations increased. Dissolved organic phosphorus showed little seasonal variation and formed a smaller fraction of the total dissolved pool compared to nitrogen. Winter inorganic nitrogen and phosphorus salinity relationships were conservative: winter organic nitrogen and phosphorus salinity relationships were not conservative. In the winter, dissolved organic nitrogen and phosphorus formed a significant fraction of the total dissolved pool.

Introduction

Dissolved organic nitrogen (DON) and dissolved organic phosphorus (DOP), derived from excretory and decay products, are important fractions of marine nutrient pools, especially in the euphotic zone. They possibly supply the summer nitrogen (N) and phosphorus (P) requirement of micro-organisms at a time when inorganic nutrients are scarce. Drivers for determining the stoichiometry and total quantities of accessible dissolved nutrients when considering phytoplankton and nutrient dynamics in the marine environment are concerns about eutrophication, real or perceived; EC directives such as the Urban Waste Water Treatment Directive and the Nitrate Directive; international agreements such as the OSPAR Convention.

The Irish Sea (Fig. 1) is a semi-enclosed sea less than one-tenth the area of the North Sea. It covers an area north of a line across the St George's Channel (~52° N) in the south and south of a line across the North Channel (~55° N) in the north. Its characteristics have been discussed by, for example, Simpson (1971), Pingree & Griffiths (1978), Dickson & Boelens (1988), Irish Sea Study Group (1990) and Hill et al. (1997). With hydrodynamic residence times of up to 1 year in some areas, the Irish Sea is vulnerable to contamination. The English coast is a major source of riverine dissolved nitrogen and phosphorus including anthropogenic inputs.

In the Irish Sea, stretches of the English and Welsh coastline are hypernutrified and there is the possibility of future eutrophication problems in these areas (Kennington et al., 1999). Waters in the central and western Irish Sea cannot be classified as hypernutrified (Shammon et al., 1999), though a time-series from a central Irish Sea site suggests that levels of concentrations of dissolved inorganic nitrogen and phosphorus have approximately doubled over the last half-century (Allen et al., 1998).

Presented here are the 1997 and 1998 spatial distributions and partitioning of winter and summer dissolved N and P using water column profiles from nine routine monitoring stations along an east–west transect across the Irish Sea.

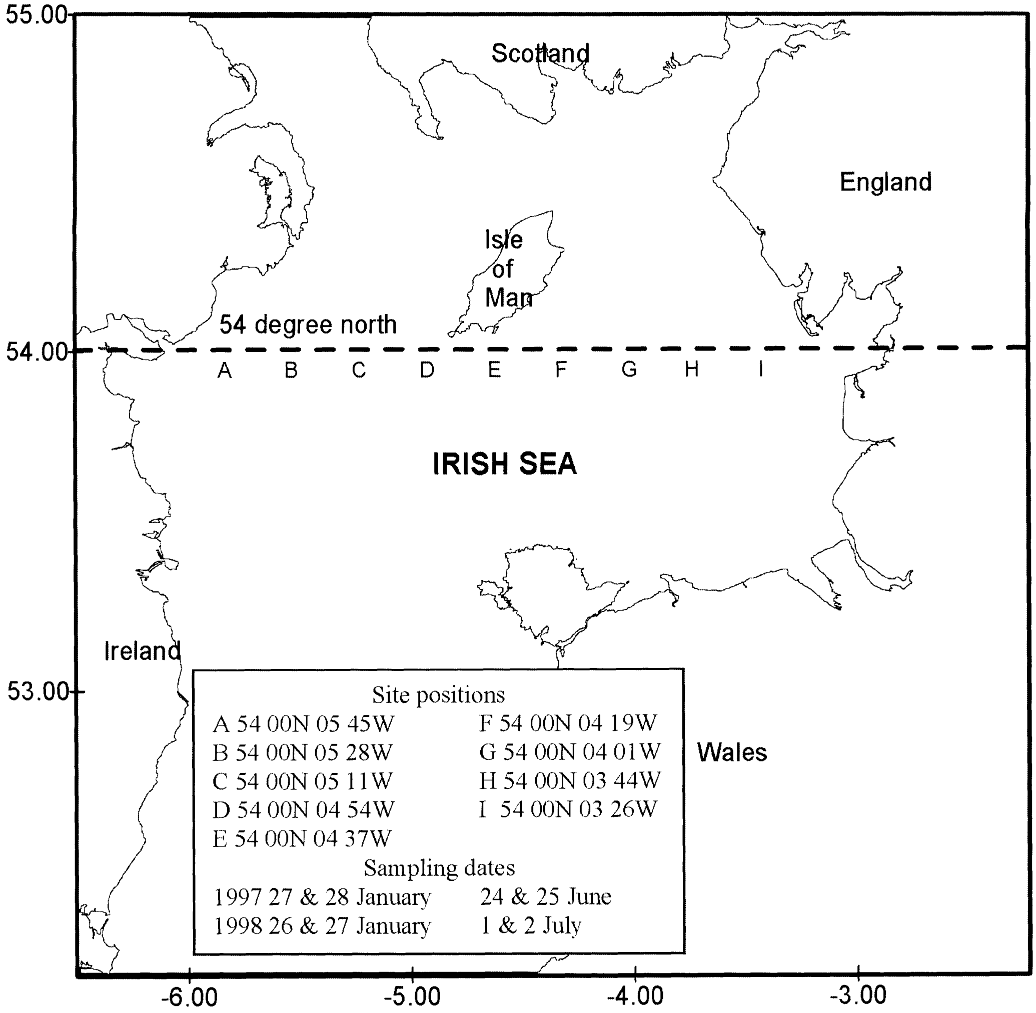

Figure 1. Position of sampling sites.

Sampling and methods

Measurements and samples were taken in the Irish Sea between Carlingford Lough (Ireland) and Morecambe Bay (England) at nine stations approximately 10 nautical miles apart along a transect that co-incided with the 54° N parallel (Fig. 1). The sites were visited twice yearly, winter and summer. Sampling was performed aboard the RV Roagan (University of Liverpool) and was part of an ongoing monitoring programme at Port Erin Marine Laboratory (PEML) funded by the Isle of Man Government.

Physical structure of the area was elucidated using a Seacat SBE 19 (Sea-Bird Electronics Inc., Bellevue, Washington, U.S.A.) CTD profiler. CTD data were averaged into 1-m depth bands. Stratification parameters were calculated using the formulae of Simpson et al. (1978). Water samples were taken at 20 m depth intervals using IOS type sample bottles or a Sea-Bird Electronics rosette sampler.

Samples were filtered using Whatman membrane filters, pore size 0.45 μM, mounted on Nalgene filter holders of 250 ml capacity, transferred to acid washed HDPE bottles and immediately frozen (−18 °C±2 °C) on board ship for analysis within 4 weeks. Unfiltered samples were also taken and frozen for the estimation of particulate bound N and P, however, it later became apparent that the method of sampling and storage was inappropriate (Nausch, 2000).

Inorganic nutrients, soluble reactive phosphate (SRP) and total oxidised nitrogen, nitrate plus nitrite, (N+N), were analysed with a RFA^{TM} Alpkem RFA/2 autoanalyser using the standard colorimetric methodology recommended by the manufacturer (RFA^{TM}, 1986–1990) and the chemistry outlined in Parsons et al. (1984).

Dissolved nitrogen and phosphorus compounds were converted off-line to total dissolved phosphorus (TDP) as SRP and total dissolved nitrogen (TDN) as nitrate by oxidation with di-potassium peroxodisulphate (potassium persulphate). Reaction vessels were PTFE bottles of 60 ml capacity. Before use these were cleaned with 10% Decon®, followed by concentrated sulphuric acid. Cleaned vessels were stored filled with de-ionised (UHQ) water. Immediately prior to use the vessels were pre-cleaned with the relevant oxidising reagent and autoclaved. Inorganic standards and recoveries were diluted using artificial seawater (Grasshoff, 1976). Standard low nutrient seawater, although low in inorganic nutrients, was found to have a significant concentration of total phosphorus and nitrogen and therefore not used in the analysis of total nutrients. Inorganic standards were supplied by Ocean Scientific International Limited. Analytical grade reagents were used throughout. Routine recovery standards were DL-α-Glycerophosphate (di-Sodium salt. Hexahydrate) and urea, which gave recoveries of 96–102%. PEML has subscribed to QUASIMEME Laboratory Performance Studies since 1996 with acceptable results for both inorganic and total nutrients.

The oxidising reagent for phosphorus digestion was 20 g of potassium persulphate in 250 ml of dilute (4 ml l^{-1}) sulphuric acid. Forty-ml aliquots of sample were pipetted into the reaction vessels with 5 ml of (phosphorus) oxidising reagent. The vessels were loosely capped and autoclaved for 90 min at 15 psi, 125 °C (Menzel & Corwin, 1965; Koroleff, 1976; Ridal & Moore, 1990). Dissolved nitrogen compounds were converted to total dissolved nitrogen (TDN) as nitrate, with alkaline potassium peroxodisulphate, following a method similar to that outlined in Solorzano & Sharpe (1980). The heating time was increased to ensure full decomposition and reduce the risk of oxidation of the cadmium coil of the autoanalyser. The oxidations were followed by analysis for SRP and N+N using the autoanalyser.

For this study, dissolved organic nitrogen (DON) was calculated as the difference between TDN and N+N: dissolved organic phosphorus (DOP) was calculated as the difference between TDP and SRP. DON included ammoniacal nitrogen, concentrations of which were low during both winter and summer, and as such were considered prone to error (Aminot et al., 1997). The nitrite fraction of N+N was generally less than 0.1 μM in the winter and 0.2 μM in the summer.

Contour diagrams were produced using Surfer for Windows, version 6 (Golden Software). Statistical data were generated using Microsoft Excel and Grapher for Windows, version 2 (Golden Software).

Results

Physical structure of the water column

Salinity (Figs 2b, d and 3b, d) in general exhibited an east (~33.4) to west (~34.5) horizontal increase, with less variation in the central and western Irish Sea. Distribution patterns were similar on the four sampling occasions. In Morecambe Bay, at the eastern end of the transect, 1998 winter salinities were lower (32.3) compared to 1997 (33.6). There were significant linear correlations between salinity and temperature. This correlation was strongly positive in winter; r^2 (df 28) was 0.90 and 0.87 for winter 1997 and 1998, respectively. Correlation was negative in summer; r^2 (df 28) was 0.51 and 0.44 for summer 1997 and 1998, respectively. Winter temperature (Fig. 2a, c) in general exhibited an east to west horizontal increase, with less variation in the central and western Irish Sea. Winter temperatures were higher in 1998 compared to 1997. During the summer there was a general east to west horizontal decrease in temperature (Fig. 3a, c) with vertical stratification and a well-developed thermocline in the western Irish Sea. Stratification parameters are presented in Table 1. The degree of stratification was greater in summer 1998 compared to 1997. The winter water column was generally vertically well mixed. In January 1998, the water column at the eastern end of the transect was intermediate in nature with some evidence of haline stratification. Summer temperatures were higher in 1997 compared to 1998.

Salinity-nutrient associations

Regression data for winter dissolved nitrogen and salinity are presented in Figure 4a, b and Table 2. There were strong negative linear correlations between winter N+N concentration and salinity for individual years and also combined data. There was no significant linear correlation between winter DON concentration and salinity for 1998; there was a weak, but significant correlation between 1997 DON concentration and salinity.

Table 1. Stratification parameters after Simpson (1971). (**bold** – vertically stratified waters: *italics* – waters intermediate in nature: normal – vertically mixed waters). (V – stratification parameter: ΔS – vertical salinity difference: ΔT – vertical temperature difference)

Station	Position	Nominal depth	January 1997			January 1998		
			V	ΔS	ΔT	V	ΔS	ΔT
A	05 45 W	45	−0.7	−0.22	0.07	3.6	−0.15	−0.17
B	05 28 W	106	*14.6*	−0.07	0.08	*12.5*	−0.2	−1.17
C	05 11 W	62	0.4	−0.01	−0.01	10.7	−0.55	0.59
D	05 54 W	46	1.9	−0.13	−0.01	3.4	−0.01	−0.69
E	04 37 W	42	−0.8	−0.03	0.01	8.8	−0.25	−0.08
F	04 19 W	40	−0.1	0.01	0.02	0.0	0.01	−0.02
G	04 01 W	42	−1.3	0.03	0.04	1.4	0.09	0.00
H	03 44 W	34	−1.5	0.08	0.16	9.7	−0.43	−0.88
I	03 26 W	22	−1.5	0.04	0.04	*12.4*	−1.11	−0.62
			June 1997			July 1998		
A	05 45 W	45	*15.5*	−0.05	1.57	**26.8**	0.31	3.05
B	05 28 W	106	**55.0**	−0.19	2.54	**73.5**	−0.28	4.79
C	05 11 W	62	*18.2*	−0.07	1.37	**37.4**	−0.14	2.64
D	05 54 W	46	7.0	0.01	0.10	5.8	−0.24	0.56
E	04 37 W	42	−1.0	0.03	0.08	1.4	−0.02	0.43
F	04 19 W	40	1.8	−0.13	0.10	1.2	0.07	0.69
G	04 01 W	42	8	−0.21	0.29	5	0.36	0.69
H	03 44 W	34	3.8	−0.11	0.41	2.4	0.34	1.40
I	03 26 W	22	6.8	−0.21	0.69	3.6	0.10	1.12

Table 2. Regression data for dissolved nutrient–salinity associations

Season	Year	Variable	Slope	Intercept	r^2	Number of data points	Average N or P μM	Average salinity
Winter	1997	N+N	−6.95	247.0	0.93	30	7.7	34.5
Winter	1998	N+N	−8.00	286.8	0.97	30	9.8	34.6
Winter	all data	N+N	−7.35	262.6	0.82	60	8.7	34.5
Winter	1997	DON	−3.80	135.3	0.39	29	5.8	34.4
Winter	1998	DON	−1.53	58.3	0.01	25	5.0	34.8
Winter	1997	SRP	−0.86	30.6	0.98	30	0.8	34.5
Winter	1998	SRP	−0.41	14.8	0.95	30	0.7	34.6
Winter	all data	SRP	−0.47	17.2	0.86	60	0.7	34.5
Winter	1997	DOP	0.08	−2.7	0.10	29	0.1	34.5
Winter	1998	DOP	0.03	−0.8	0.07	24	0.1	34.6
Summer	1997	N+N	2.66	−89.73	0.19	30	1.9	34.4
Summer	1998	N+N	1.89	−64.01	0.10	29	1.2	34.4
Summer	1997	DON	−0.68	31.84	0.03	30	8.4	34.4
Summer	1998	DON	2.31	−67.17	0.04	30	12.3	34.4
Summer	1997	SRP	0.01	0.77	0.00	30	0.3	34.4
Summer	1998	SRP	0.14	−4.83	0.07	29	0.2	34.4
Summer	1997	DOP	−0.02	1.00	0.01	30	0.2	34.4
Summer	1998	DOP	−0.17	6.09	0.41	30	0.2	34.4

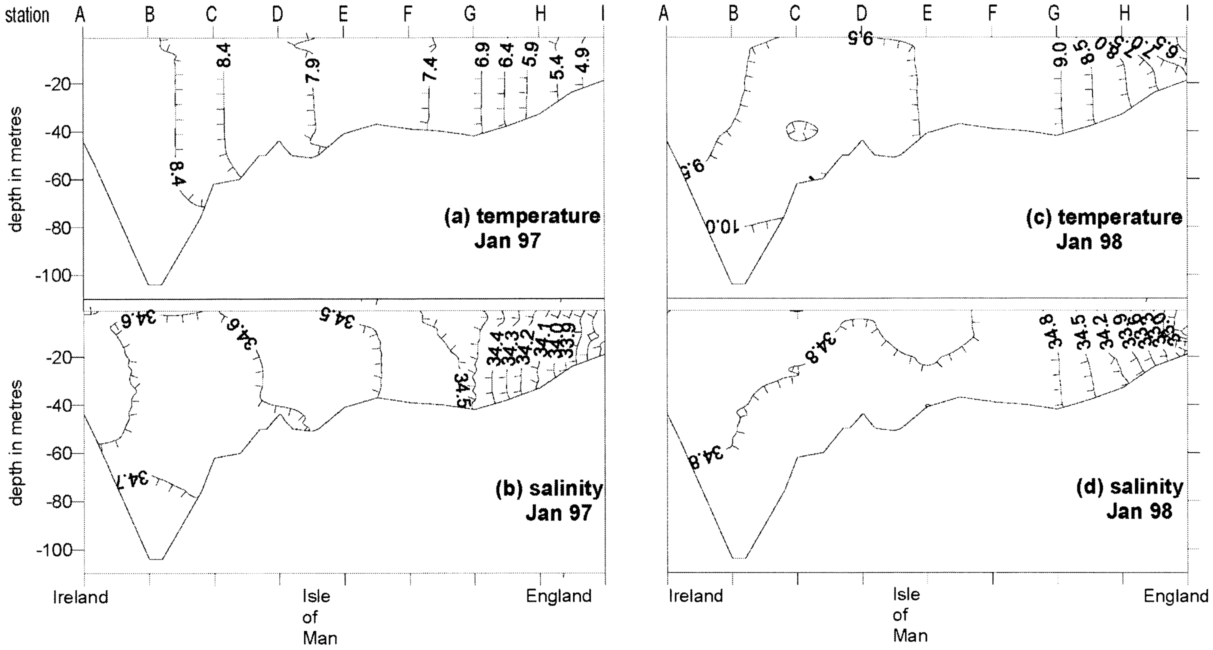

Figure 2. Profiles of winter temperature and salinity. The contour intervals are (a) and (c) 0.5 °C, (b) 0.1 and (d) 0.3. Hachures and labels are orientated uphill.

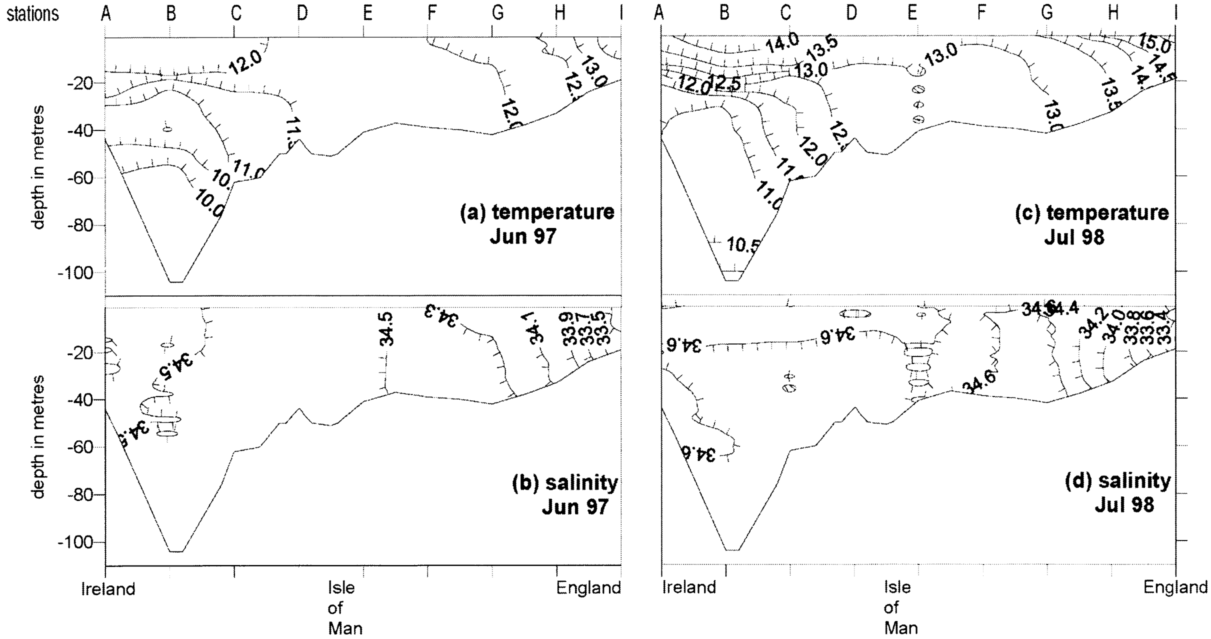

Figure 3. Profiles of summer temperature and salinity. The contour intervals are (a) 1.0 °C, (c) 0.5 °C, (b) and (d) 0.2. Hachures and labels are orientated uphill.

Regression analyses for winter dissolved phosphorus and salinity are presented in Figure 4c, d and Table 2. There were strong negative linear correlations between winter SRP and salinity for individual years and also combined data. There were no significant linear correlations between winter DOP and salinity.

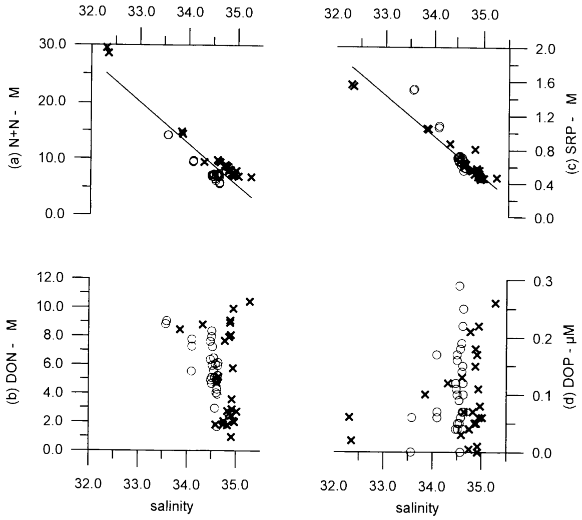

Figure 4. Winter dissolved nitrogen and phosphorus–salinity relationships. (a) N+N vs salinity, (b) DON vs salinity, (c) SRP vs salinity and (d) DOP vs salinity. (o – 1997 data, x – 1998 data, best fit all data - —). Regression data are presented in Table 2.

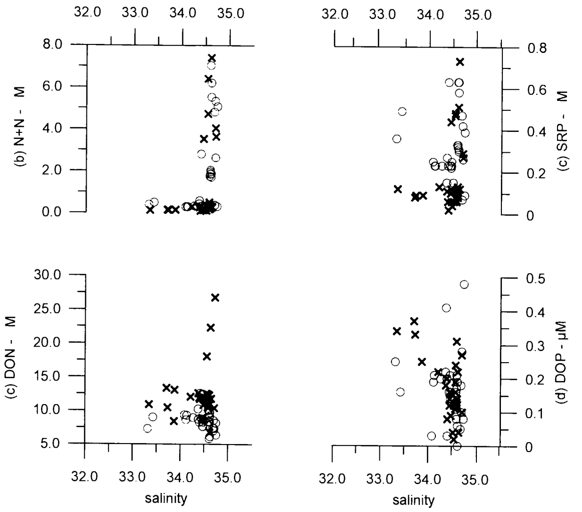

Figure 5. Summer dissolved nitrogen and phosphorus–salinity relationships. (a) N+N vs salinity, (b) DON vs salinity, (c) SRP vs salinity and (d) DOP vs salinity. (o – 1997 data, x – 1998 data all data). Regression data is presented in Table 2.

Regression data for summer dissolved nitrogen and salinity are presented in Figure 5a, b and Table 2. There was a very weak but just significant linear correlation between N+N concentration and salinity for summer 1997 but not in 1998. There were no significant linear correlations between summer DON concentration and salinity.

Regression data for summer dissolved phosphorus and salinity are presented in Figure 5c, d and Table 2. There were no significant linear correlations between SRP concentration and salinity. There was no significant linear correlation between DOP concentration and salinity for summer 1997; there was a significant, negative linear correlation between DOP concentration and salinity for summer 1998.

Winter nitrogen and phosphorus

Winter concentrations of dissolved nitrogen are presented as contour maps in Figure 6a–d. Highest concentrations of N+N were found at the eastern end of the transect, in Morecambe Bay, 14.26 μM and 29.50 μM for 1997 and 1998, respectively. At the eastern end of the transect, the 1998 winter N+N fraction of the TDN pool was >90%. In the eastern Irish Sea, there was a horizontal east to west decrease in concentration of N+N. Concentration in the central and western Irish Sea was broadly similar and in the range of 6–8 μM. Overall DON formed ~43% and ~32% of the winter TDN pool for 1997 and 1998, respectively, with concentrations up to 10 μM. Mean winter DON concentration was 5.8 μM±0.6 μM and 5.0 μM±1.22 μM for 1997 and 1998, respectively. Spatial patterns, across the Irish Sea, for the winter distributions of DON were less discernible compared to N+N.

Winter concentrations of dissolved phosphorus are presented as contour maps in Figure 7a–d. Highest concentrations of SRP were found at the eastern end of the transect, 1.50 μM and 1.56 μM for 1997 and 1998, respectively. In the eastern Irish Sea, there was a horizontal east to west decrease in concentration. Concentration in the central and western Irish Sea was broadly similar and in the range 0.5–0.6 μM. DOP formed ~11% and ~18% of the winter TDP pool in 1997 and 1998, respectively with concentrations up to 0.3 μM. DOP concentrations were 0.12 μM±0.03 μM and 0.08 μM±0.03 μM for 1997 and 1998, respectively. As with DON, spatial patterns for winter distributions of DOP were less discernible compared to SRP.

Summer nitrogen and phosphorus

There was a winter to summer decrease in concentration of N+N and SRP, and also of TDN and TDP which was accompanied by an increase in DON concentration and a less discernible increase in DOP concentration. Summer concentrations of dissolved nitrogen are presented as contour maps in Figure 8a–d. Concentrations of N+N in 1997 in the central Irish Sea were up to 1.5 μM, but in the eastern Irish Sea,

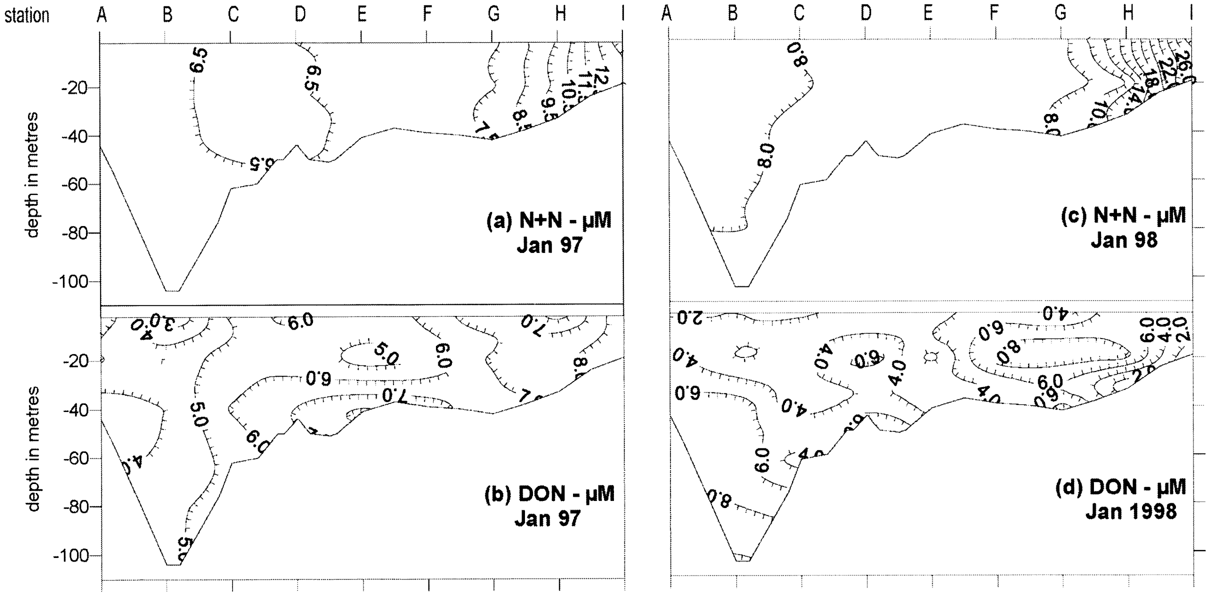

Figure 6. Profiles of winter N+N and DON. The contour intervals are (a) and (b) 1.0 μM, (c) and (d) 2.0 μM. Hachures and labels are orientated uphill.

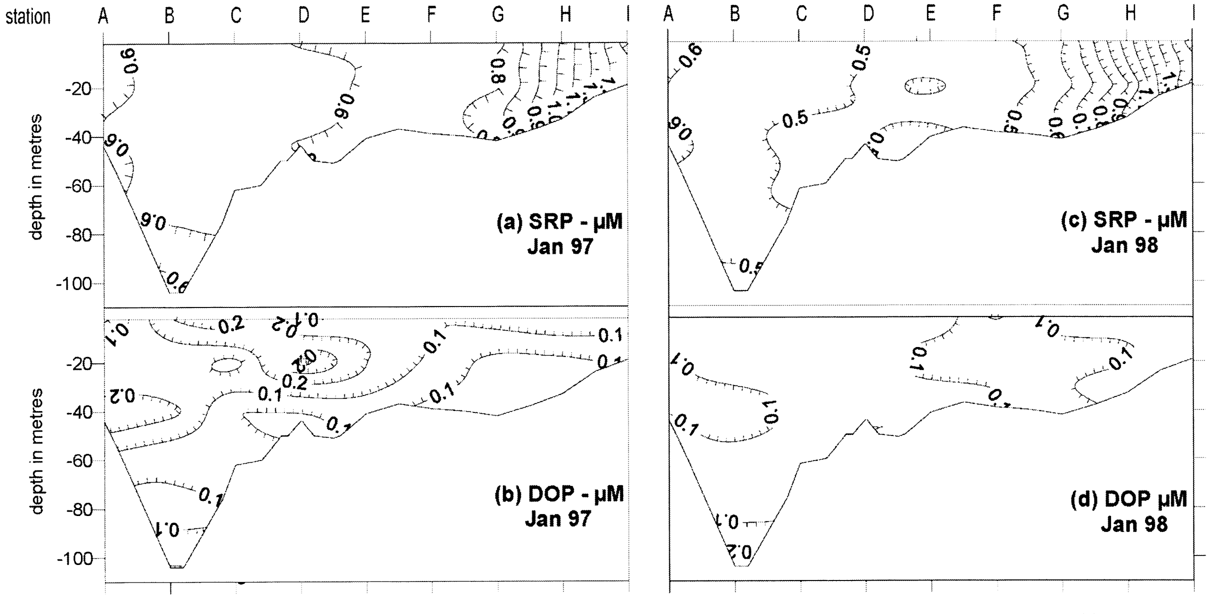

Figure 7. Profiles of winter SRP and DOP. The contour intervals are 0.1 μM. Hachures and labels are orientated uphill.

and above the themocline in the euphotic zone of the western Irish Sea, concentrations were generally below 1.0 μM. Below the thermocline, in the deeper waters of the western Irish Sea, there was a vertical increase in N+N concentration to 7.0 μM (1997) and 7.5 μM (1998). DON formed ~90% of the summer TDN pool; below the thermocline DON formed <60% of the TDN pool. There was some evidence of a decrease in DON concentration in waters below 70–80 m (Fig. 8b, d). In 1997, mean summer DON concentration was 8.4 μM±0.5 μM, with maximum concentrations up to 12.6 μM, and in 1998 mean summer DON concentration was 12.3 μM±0.9 μM with maximum concentrations up to 18.0 μM. Spatial patterns for the summer distribution of DON were not easily discernible.

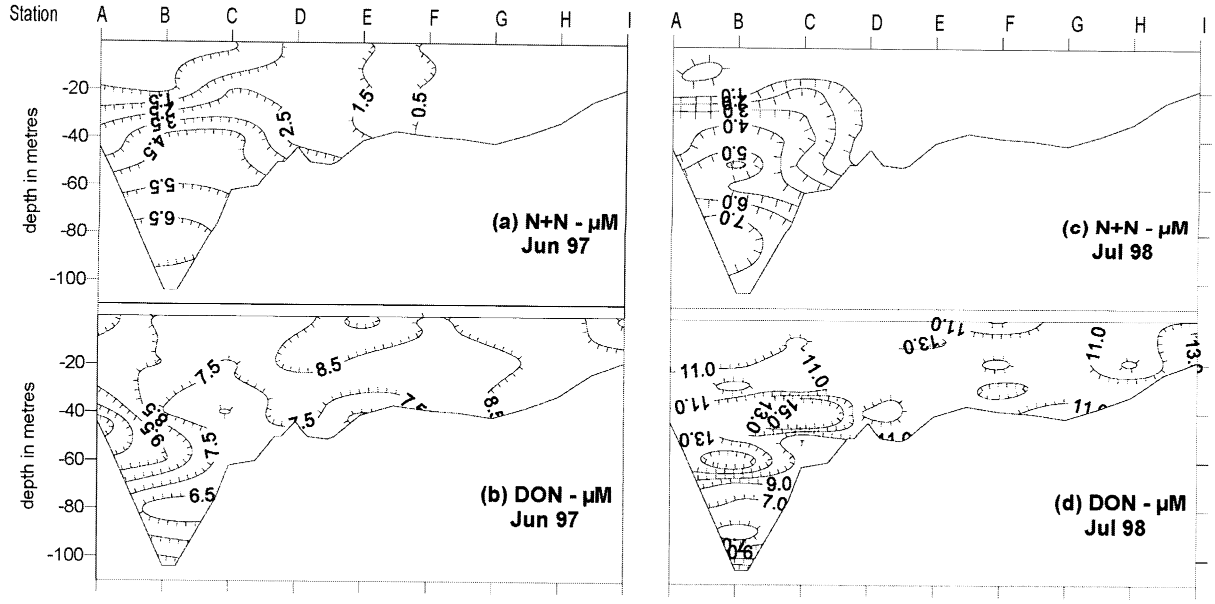

Figure 8. Profiles of summer N+N and DON. The contour intervals are (a), (b) and (c) 1.0 μM and (d) 2.0 μM. Hachures and labels are orientated uphill.

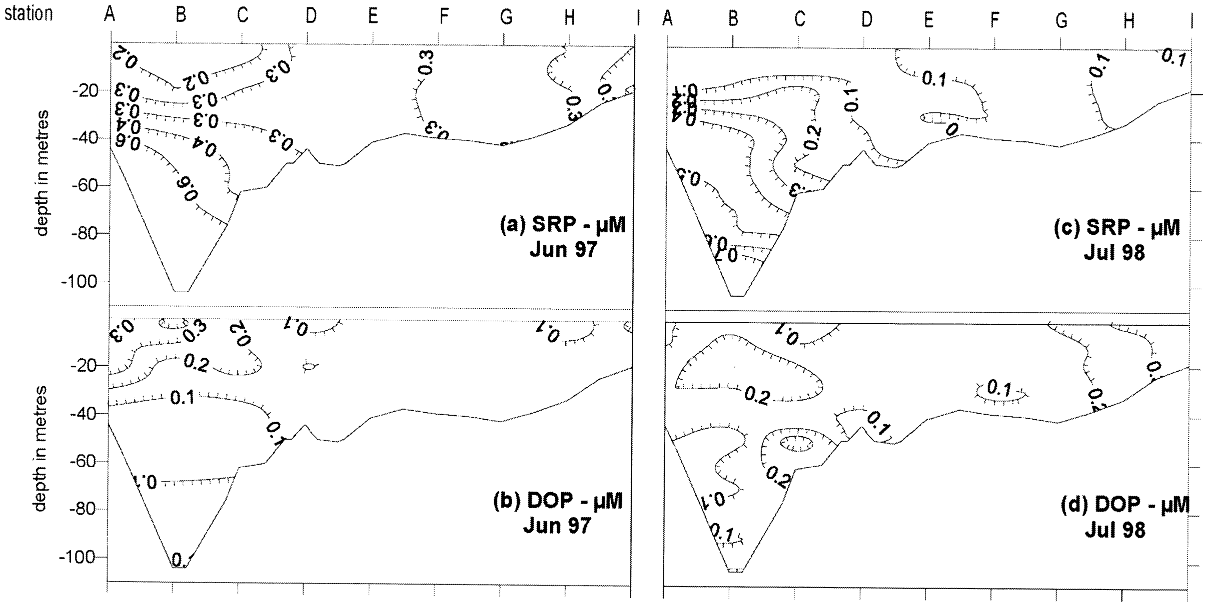

Figure 9. Profiles of summer SRP and DOP. The contour intervals are (a) 0.2 μM and (b), (c) and (d) 0.1 μM. Hachures and labels are orientated uphill.

Summer concentrations of dissolved phosphorus are presented as contour maps in Figure 9a–d. In the eastern Irish Sea, and above the themocline in the euphotic zone of the western Irish Sea, concentrations of SRP were in the region of 0.3 μM (1997) and 0.1 μM (1998). Below the thermocline, in the deeper waters of the western Irish Sea, there was a vertical increase in SRP concentration to 0.6 μM (1997) and 0.7 μM (1998). DOP formed ±54% and ±36% of the summer TDP pool for 1997 and 1998, respectively. Below the thermocline this figure fell to <25%. Mean summer DOP concentration was 0.17 μM±0.04 μM for both years. Maximum concentrations were 0.38 μM and 0.48 μM for 1997 and 1998, respectively. Spatial pat-

Table 3. Regression data for dissolved nitrogen–phosphorus associations

Season	Year	Variables	Slope	Intercept	r^2	Number of data points
Winter	1997	N+N:SRP	7.96	1.67	0.93	30
Winter	1998	N+N:SRP	18.67	−2.21	0.93	30
Winter	1997	TDN:TDP	12.20	2.89	0.73	28
Winter	1998	TDN:TDP	16.00	2.97	0.74	30
Winter	1997	DON:DOP	−13.61	7.29	0.32	26
Winter	1998	DON:DOP	23.89	3.06	0.26	22
Summer	1997	N+N:SRP	11.02	−1.66	0.64	30
Summer	1998	N+N:SRP	11.70	−0.75	0.90	30
Summer	1997	TDN:TDP	8.91	5.88	0.30	30
Summer	1998	TDN:TDP	12.07	8.52	0.52	27
Summer	1997	DON:DOP	−2.67	8.86	0.04	30
Summer	1998	DON:DOP	0.62	11.47	0.00	27

terns for the summer distributions of DOP were not easily discernible.

Nitrogen and phosphorus ratios

The Redfield ratio for seawater is given by the slope of a regression line to fit the N vs. P data. The traditional Redfield N:P ratio is 16:1. Regression data of N+N:SRP, TDN:TDP and DON:DOP for winter and summer are presented in Figure 10a–f and Table 3.

Slopes for linear regressions, i.e. Redfield ratios, between winter N+N and SRP were 8.0 and 18.7 1997 and 1998, respectively. TDN and TDP slopes were 12.2 and 16.0 for 1997 and 1998, respectively. The 1997 winter TDN:TDP values were generally below the 'Redfield line', whilst 1998 values were generally above the 'Redfield line'. Winter N+N:SRP values for both years were generally below the 'Redfield line'. Compared to TDN:TDP and N+N:SRP values, winter DON:DOP values were scattered. Linearity, however, was significant with slopes of −13.6 and 23.9 for 1997 and 1998, respectively, with values generally above the 'Redfield line'.

Slopes for linear regressions between summer TDN and TDP were 8.9 in 1997 and 12.4 in 1998. Slopes for linear regressions between summer N+N and SRP were 11.0 and 11.7 for 1997 and 1998, respectively. Summer N+N:SRP values fell below the 'Redfield line' whilst TDN:TDP values for both years were mostly above the 'Redfield line'. There was no significant linearity for DON:DOP data.

Winter mean arithmetic quotients for N+N:SRP were 10.4±0.4 (1997), 15±0.6 (1998), TDN:TDP

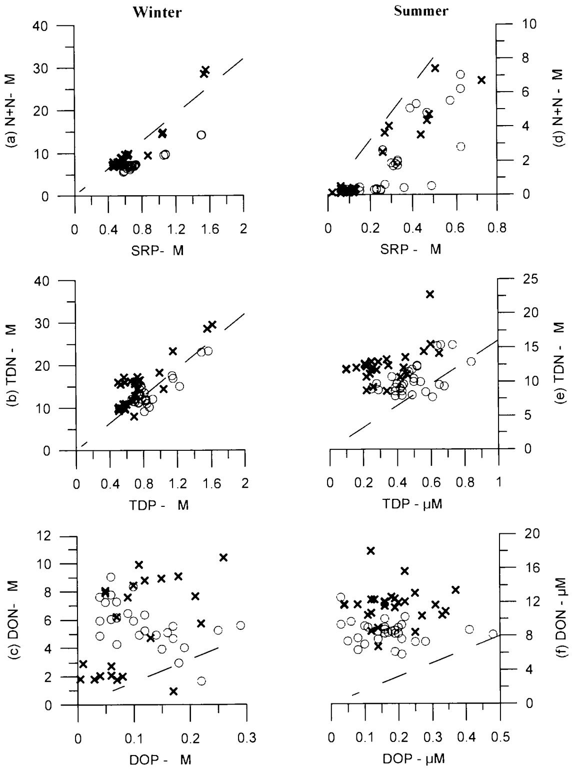

Figure 10. Nitrogen–phosphorus linear relationships. (a) winter N+N vs SRP (b) winter TDN vs TDP, (c) winter DON vs DOP, (d) summer N+N vs SRP, (e) summer TDN vs TDP and (f) summer DON vs DOP. (o – 1997 data, x – 1998 data, 'Redfield line' - —). Regression data are presented in Table 2.

15.8±0.8 (1997), 20.6±1.6 (1998) and DON:DOP 70.5±19.7 (1997), 85.8±36.6. Summer mean arithmetic quotients for N+N:SRP were 4.8±1.4 (1997), 5.4±1.2 (1998), TDN:TDP 21.9±2.0 (1997), 45.8±6.7 (1998) and DON:DOP 78.4±30.67 (1997), 97.8±20.8 (1998).

Discussion

The bulk of riverine input into the Irish Sea is via the rivers of the Solway Firth, Mersey, Ribble and Eden. Nitrogen and phosphorus inputs to the eastern Irish Sea via these rivers are estimated to be around 77% and 49%, respectively (Kennington et al., 1999). Riverine input to the western Irish Sea is considerably less. Concentrations of inorganic nutrients entering via the St George's Channel in the south are close to concentrations currently encountered in the central and western Irish Sea. These observations were reflected in the concentrations and distributions of inorganic nutrients detected in this study, which were in line with those observed for recent years. In Morecambe Bay winter inorganic nutrient concentrations were such that these waters could be considered hypernutrified under guidelines set out by the Comprehensive Studies Task Team (CSST, 1997). The greater N+N concentrations of 1998 can be explained by prevailing meteorological conditions. Rainfall data for the Irish Sea from Ronaldway Meteorological Office, Isle of Man for the 2 months preceding winter sampling show that rainfall totalled 70 mm in 1997, but 182 mm in 1998. Increased precipitation would increase riverine input. N+N also represents an important fraction of rainwater (Preston et al., 2000).

Along the transect, the winter relationship between dissolved inorganic nutrients and salinity was conservative, indicating that the main influence on dissolved inorganic N and P was riverine. Total dissolved nutrients and salinity exhibited a lesser linearity compared to inorganic nutrients. Although conservative, the TDN and TDP pools were influenced by N and P sources other than riverine, this influence being the dissolved organic fraction of the total dissolved pool. Winter dissolved organic nutrients and salinity exhibited little or no linearity; dissolved organic nutrients were not conservative. Rivers are probably of minor importance as sources of dissolved organic matter (DOM) in the marine environment (Williams, 1975), with *in situ* release by organic decay or excretion by phytoplankton likely to represent a considerable source of DOM. The atmosphere is also a significant source of DOM (Williams, 1975). Summer dissolved nitrogen and phosphorus fractions exhibited little or no linearity with salinity. Processes other than riverine predominantly influenced concentrations, these processes being presumably related to phytoplankton uptake and release mechanisms.

The seasonal inverse relationship between inorganic and organic N, observed in this study, is not unusual and has been noted by other workers (e.g. Jackson & Williams, 1985). The small decrease observed in the summer TDN and TDP pools represents a net export of N and P to the macrobiota at this time of year. Below the euphotic zone and summer thermocline the observed increase in the inorganic fraction of the summer TDN and TDP pools was an effect of inorganic nutrient regeneration processes becoming more dominant at these depths. That DOP formed a smaller fraction of the TDP pool compared to DON and the TDN pool, both in winter and in summer, may be a function of the differing chemistries of the phosphorus and nitrogen cycles and a more rapid turnover of DOP compared to DON. A smaller more labile DOP pool possibly explains the absence of an observed inverse inorganic–organic P relationship for this study. Banoub & Williams (1973) observed evidence of a summer DOP increase in some years but not others, at station E_1 in the English Channel, reasons for which were not understood. Other workers (e.g. Jackson & Williams, 1985) have observed a modest inverse relationship between inorganic and organic P.

The size and concentration of the inorganic and organic fractions of the winter and summer TDN and TDP pools represents a hydrochemical balance. The chemical variation of the DON and DOP pools is subject to seasonal variation. Phytoplankton are the main producers of summer DOM. The composition of this DOM will vary with the species composition of phytoplankton communities, which in turn will vary with physical variables such as hydrological and meteorological conditions. The Irish Sea has a range of hydrographic regimes within a small area, and the distribution and the species composition of phytoplankton communities within these regimes is also variable (Kennington et al., 1999). The spatial and temporal chemical composition of the reactive fraction of summer DOM is unlikely to be consistent. The labile fractions of summer DON and DOP pool could have the capacity to act as sources of N and P for phytoplankton, though within the study area it cannot be assumed that they are sources of N and

P for phytoplankton. It is probable that many species of phytoplankton can utilise a variety of nitrogen sources at environmental concentrations. Variations in the relative fractions of different forms of nitrogen between environments, or at different times in a particular environment, may be more important as an influence upon the species composition of a population and species succession, than in determining total productivity (Raymont, 1980). The significant pools of winter DON and DOP observed in this study may represent a more chemically recalcitrant fraction of DOM.

Results from the Pacific (Jackson & William, 1985) showed that inclusion of DON and DOP with nitrate and phosphate to form TDN and TDP improved the Redfield argument linking organism N:P values with nitrate:phosphate values. One caveat was that at phosphate concentrations $<0.8\ \mu$M, most nitrate:phosphate values fell below the Redfield regression line. Conclusions from this study are uncertain. For the study area in winter, most SRP concentrations were $<0.8\ \mu$M. Most TDP values were also $<0.8\ \mu$M. The 1997 TDN:TDP values were mostly below the 'Redfield line' giving an overall ratio of 12, however, in 1998 TDN:TDP values were mostly above the 'Redfield line' giving an overall ratio of 16, the classic ratio. It is possible that the range of hydrodynamic regimes within the Irish Sea may make it unamenable to analysis using the Redfield relationship.

The accepted convention amongst marine biologists is that nitrogen, rather than phosphorus, is the limiting factor for primary production in coastal waters (Hecky & Kilham, 1988). Results from this study neither confirm nor refute this paradigm. Redfield et al. (1963) concluded that a N:P ratio which is on average 7:1 is optimal for primary production, and if N or P is present in a smaller degree then that is the one that is limiting. Redfield (1958) had previously concluded that phosphorus limits primary production in the sea. Alternatively, Forsberg et al. (1978) found that if the N:P ratio was <10 then nitrogen limits primary production and if the ratio is >17 then phosphorus limits primary production. Hanninen et al. (2000) notes that conclusions from studies in which nutrient ratios have been used as a measure of limiting factors have been extensively criticised as interpretative.

Implicit in this study is the presumption that the measurements of TDN and TDP are accurate. The method of evaluating total nitrogen and phosphorus used in this study is a standard, widely applied technique and performance and results are in line with other workers (QUASIMEME, 2000). However, Suzuki et al. (1985) and Sugimura & Suzuki (1988) questioned measurements of dissolved organic matter (DOM). It was suggested that the TDN pool in the Pacific Ocean was at least three times greater than previously estimated by standard methods of analysis. Williams & Druffel (1988) and Karl et al. (1993) emphasised that these, as yet unsubstantiated, claims need to be fully evaluated by independent investigators. Ridal & Moore (1990) suggested that the persulphate technique may be in error by a factor $\sim$1.25–1.5 for open ocean surface waters, but adequately measures the DOP content in other seawaters. The presence of a fraction of the DOM pool that is labile to the marine microbial community but refractory to chemical methods cannot be ruled out.

Acknowledgements

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Long-term changes in Wadden Sea nutrient cycles: importance of organic matter import from the North Sea

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Key words: Wadden Sea, eutrophication, long-term changes, nitrogen cycle

Abstract

The Wadden Sea is a shallow tidal area along the North Sea coast of The Netherlands, Germany and Denmark. The area is strongly influenced by rivers, the most important of which are the rivers Rhine, Meuse and Elbe. Due to the increased nutrient load into the coastal zone the primary production in the Wadden Sea almost tripled during the past few decades. A conceptual model is presented that links nitrogen input (mainly nitrate) via Rhine and Meuse with the annual nitrogen cycle within the Wadden Sea. Three essential steps in the model are: (1) nitrogen limits the primary production in the coastal zone, (2) a proportional part of the primary produced organic matter is transported into the Wadden Sea and (3) the imported organic matter is remineralized within the Wadden Sea and supports the local productivity by nitrogen turn-over. The conceptual model predicts that during years with a high nutrient load more organic matter is produced in the coastal zone and more organic matter is transported into and remineralized within the Wadden Sea than during years with low nutrient loads. As a proxy for the remineralisation intensity ammonium plus nitrite concentrations in autumn were used. Based on monitoring data from the Dutch Wadden Sea (1977–1997) the above mentioned model was statistically tested. In autumn, however, a significant correlation was found between autumn values of ammonium and nitrite and river input of nitrogen during the previous winter, spring and summer. The analysis supports that in years with a high riverine nitrogen load more organic matter is remineralized within the Wadden Sea than in years with a low nitrogen load. A comparison with older data from 1960 to 1961 suggests that the remineralisation intensity in the Wadden Sea has increased by a factor of two to three. This is not reflected by a two to three-fold increase in riverine nitrogen load from 1960 to present. It is suggested that the increased remineralisation rates in the Dutch Wadden Sea between the 1960s and the 1980s/1990s are largely caused by an increased nitrogen flux through the Channel and the Strait of Dover and by an increased atmospheric nitrogen input.

Introduction

During the past century, nutrient discharge via rivers into the North Sea has increased (e.g. Van Bennekom & Wetsteijn, 1990; De Jong et al., 1999). This increase has affected both nutrient cycles, productivity and benthic communities of the Wadden Sea. De Jonge & Postma (1974) observed a change in the annual phosphorus cycle between the 1950s and the 1970s which was explained by an enhanced import of organic matter from the North Sea and subsequent remineralisation in the Wadden Sea. During the past decades, a two- to three-fold increase in phytoplankton and phytobenthos primary production was observed (Cadée, 1984; Cadée & Hegeman, 1993; de Jonge et al., 1996; Asmus et al., 1998a). Also the biomass of macrobenthos has increased in at least in some parts of the Wadden Sea (Beukema & Cadée, 1997; Asmus et al., 1998b) while also the meat content of the blue mussel (*Mytilus edulis*) increased since 1960 (de

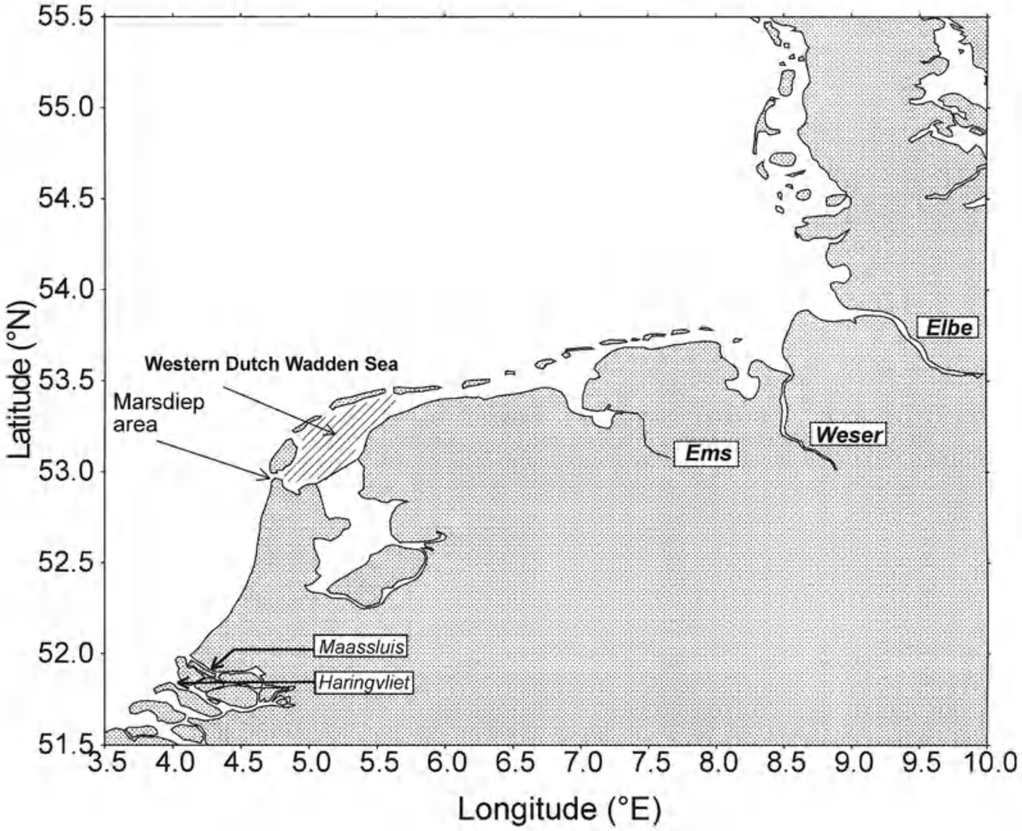

Figure 1. Map of the Wadden Sea with the main subareas used in the data analysis.

Jonge et al., 1996). Finally, within the Wadden Sea a shift towards small opportunistic species was observed (Essink et al., 1998).

Several explanations have been put forward to relate the eutrophication of the Wadden Sea with an increased nutrient loading of the Wadden Sea. De Jonge (1990) found a significant correlation between changes in mean annual chlorophyll in the Marsdiep area and annual primary production (pre-1987) with the dissolved inorganic phosphate load from the IJsselmeer into the Marsdiep area and not with other nutrient components or sources. Based on more recent primary production data, Cadée & Hegeman (1993) showed that despite decreasing phosphate loads the primary production in the Wadden Sea remained high. They observed a good correlation between annual primary production and annual Rhine discharge and suggested that the nitrogen load of the river Rhine kept the phytoplankton production in the Dutch coastal zone at a high level despite decreasing phosphorus discharges. In a later paper, De Jonge (1997) reanalyzed the productivity of the Wadden Sea and stressed the importance of increased phosphorus input through the Dover Channel. He showed that despite the decreasing riverine loads in phosphorus, this element still might play a significant role in fuelling the primary production in the western Dutch Wadden Sea. However, the increased productivity of the Wadden Sea is not only a direct effect of nutrient-rich river water being advected into the area, but also, and perhaps even mainly an indirect effect due to an increased import and remineralisation of organic matter from the adjacent coastal zone (Postma, 1954; De Jonge & Postma, 1974; Van Beusekom et al., 1999). Changes in the seasonal cycle of nutrients might be used as a direct indicator of changes in organic matter import and remineralisation rates. De Jonge & Postma (1974) already explored this strategy and inferred a tripling of the organic matter import from the North Sea into the Wadden Sea from 1950 to 1971–1972. On the other hand, Helder (1974) did not observe a clear-cut change in the annual nitrogen cycle between 1960–1962 and 1971–1972 in the Dutch Wadden Sea. Since then, little attention has been given to the relation between river input and

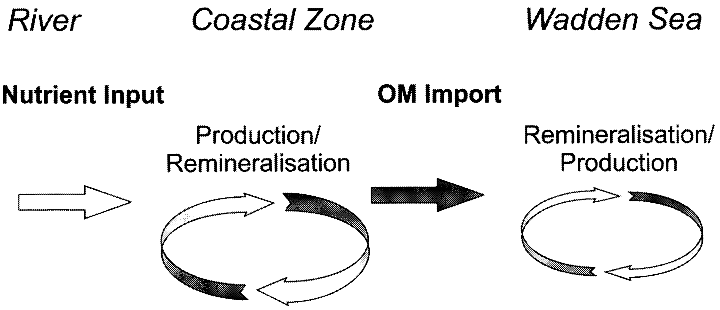

Figure 2. Conceptual Model linking nutrient input via rivers with nutrient seasonal cycles within the Wadden Sea.

the seasonal cycle of especially nitrogen within the Wadden Sea.

In this paper, long-term nitrogen data from a Dutch monitoring program in the Wadden Sea are presented. A conceptual model will be discussed that relates nitrogen input into the coastal zone with its annual cycle in the Wadden Sea. It will be shown that at present a significant relation exists between nitrogen input into the coastal zone and the intensity of the annual nitrogen cycle in the Wadden Sea. Based on a comparison with older data (1960–1962) by Postma (1966) the importance of nutrient import via the Dover Channel and of atmospheric input is stressed.

Materials and methods

Area description

The Wadden Sea is a shallow coastal area with extensive tidal flats covering about 50% of the area (Fig. 1). Its environment is very dynamic. Wind forces, water currents and water turbulence cause the formation and erosion of the typical landscape elements of the area, the tidal flats, salt marshes, sandbanks and islands. The Wadden Sea region includes an area extending from Den Helder in the Netherlands to the Skallingen peninsula in Denmark, about 500 km of coastline. It is a strip of tidal flats, sandbanks and barrier islands. On average, this strip is some 10 km wide, although in some areas it can reach a width of over 30 km. The Wadden Sea area covers approximately 13 000 km^2. Here, focus will be on the westernmost part, the western Dutch Wadden Sea.

Data base

The Wadden Sea data used in this study (1977–1997) are from the Dutch monitoring program in the western Dutch Wadden Sea. Surface water samples were collected during monthly cruises, filtered and stored frozen until analysis in the laboratory. Older data (1971–1976) are available, but their quality is questionable. The nutrient analyses were carried out with standard techniques. The data were retrieved from the Rijkswaterstaat data base DONAR. The two major rivers discharging directly into the North Sea are the rivers Rhine and Meuse. Most of the water of these rivers is discharged through the branches 'Nieuwe Waterweg' and 'Haringvliet'. For the data analysis, we used their combined nutrient loads. The river input data (1977–1993) were taken from Lenhart et al. (1996; Station 'Maassluis' at the Nieuwe Waterweg and Station 'Haringvliet') and updated with recent data (1994–1997) from Rijkswaterstaat for the same stations.

The statistical analyses have been carried out with the multiple regression module of STATISTICA (1999).

Results and discussion

Conceptual model on the relation between nutrient input into the coastal zone and the organic matter cycle in the Wadden Sea

The conceptual model behind the data analysis is as follows (Fig. 2): nutrients are discharged into the coastal zone where they enable the production of a certain amount of organic material. Part of the primary production in the coastal zone is imported into the Wadden Sea where it is remineralized. Within the Wadden Sea, primary production is enhanced by nutrients released from imported and remineralized organic matter. Below, evidence will be compiled to support the conceptual model. Three essential steps will be addressed: nitrogen limitation of the North Sea, co-variation of nutrient input and phytoplankton biomass in the North Sea and organic matter import from the North Sea into the Wadden Sea. Focus will be on nitrogen for two reasons: Firstly, it is the limiting factor for North Sea primary production (see below). Secondly, the nitrogen cycle and in particular the ammonium and nitrite cycles are directly related to the remineralisation of organic matter. Phosphorus is not considered here because, apart from biotic processes, its seasonal cycle is also strongly influenced by interactions with adsorptive compounds like iron-hydroxides (De Jonge et al., 1993b; Van Beusekom & De Jonge, 1998) hampering a straightforward interpretation of the relation between nutrient input and the annual P cycle.

Nitrogen limitation of the North Sea

It has continuously been a matter of debate, whether phosphorus or nitrogen is ultimately limiting annual primary production in the North Sea (De Jonge, 1990, 1997; Cadée & Hegeman, 1993; De Jonge et al., 1996). Evidence exists that nitrogen determines the primary production off-shore. Owens et al. (1990) and Riegman et al. (1990) showed that in the central North Sea nitrogen limits the phytoplankton productivity in summer. Hydes et al. (1999) compared the annual primary production in the North Sea south of 55° N with the annual new production based on the amount of nitrate available in March before the onset of the spring phytoplankton bloom. They included in their calculations the nitrogen load by rivers and atmosphere during the growth period. Hydes et al. (1999) calculated a five-fold annual turnover of the new production in the nitrogen-limited central North Sea as well as in the nutrient-rich Dutch and German coastal zone. Van Beusekom et al. (1999) applied a similar approach to the entire German Bight and found an annual turnover rate of 3–4. The winter nitrate concentrations and annual primary production levels range from about 7 μM NO_3 and 100 g C m^{-2} a^{-1} in the central North Sea to about 20–30 μM NO_3 and 300 g C m^{-2} a^{-1} in the German Bight (Brockmann et al., 1990; Joint & Pomroy, 1993; van Beusekom et al., 1999). The similarity in annual turnover rates in the nitrogen-rich coastal zone and in the nitrogen-poor central North Sea strongly suggests that also in the coastal zone nitrogen limits the annual primary production.

The relation between riverine nutrient loads and phytoplankton biomass in the coastal zone

Schaub & Gieskes (1991) found a positive correlation between water discharge by the river Rhine (a proxy for nutrient loads) and the phytoplankton biomass in the Dutch coastal zone. Cadée (1992) also observed a positive correlation between phytoplankton biomass in the Marsdiep and Rhine discharge. He suggested nitrogen as the causative factor. The Marsdiep is situated in the main tidal inlet of the western Dutch Wadden Sea and samples were always taken at high tide. Therefore, Cadée suggested that his time-series reflect the situation in the coastal zone. It should be noted that 50% of the fresh water in the Marsdiep area is derived from the river Rhine proper and the other 50% derived from lake IJsselmeer (e.g. De Jonge, 1990).

Import of organic matter

From a global perspective, coastal zones are heterotrophic (Smith & Hollibaugh, 1993): more organic matter is accumulated and degraded than is locally produced. Heip et al. (1995) reviewed carbon budgets from temperate coastal zones and arrived at the same conclusion. These authors suggested that import of organic matter may enhance local primary production if enough light is available.The importance of organic matter import for the Wadden Sea has already been pointed out by Postma (1954) and Postma & de Jonge (1974). Recently, Van Beusekom et al. (1999) showed that organic matter import is a general feature of the entire Wadden Sea. They concluded that about 100 g C m^{-2} a^{-1} is imported from the North Sea. This estimate is somewhat lower than 200–300 g C m^{-2} a^{-1} postulated by Postma (1984) and De Jonge & Postma (1974) for the western Dutch Wadden Sea.

The above discussion supports the link between river input and nutrient cycles in the North Sea and Wadden Sea as outlined in the conceptual model. Based on the mean seasonal cycles of phytoplankton and nitrogen compounds in the western Dutch Wadden Sea (Fig. 3) it is suggested that the amounts of ammonium and nitrite in autumn are indicators of the annual amount of organic matter remineralized in the Wadden Sea. The seasonal phytoplankton cycle shows a typical summer maximum and low values in winter. The low winter biomass is mainly due to light limitation in winter (Cadée, 1986). Ammonium and nitrite show an opposite cycle and reach minimum values in summer and maximum values in autumn. The autumn

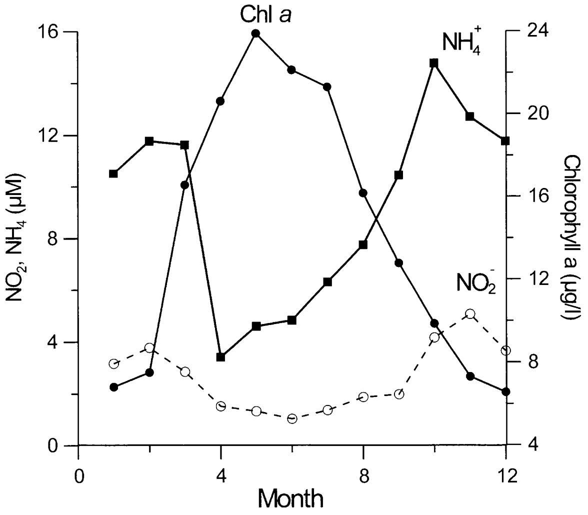

Figure 3. Mean seasonal cycle of ammonium, nitrite and chlorophyll in the western Dutch Wadden Sea.

ammonium peak occurs about 1 month earlier than the nitrite peak. This phenomenon was already observed by Postma (1966) and Helder (1974) and reflects the remineralisation sequence from organic nitrogen compounds to nitrate with ammonium and the nitrite as intermediates (Fig. 3). Despite the import and remineralisation of organic matter, the concentration of nitrite and ammonium during summer are low. This suggests that the phytoplankton is capable of removing more ammonium and nitrite than is released by remineralisation processes. However, in autumn, when the light conditions deteriorate, the phytoplankton biomass decreases and an autumn peak of nitrite and ammonium can develop.

The conceptual model predicts that during years with a high riverine nitrogen input a higher autumn peak of ammonium and nitrite can develop than during years with a low nitrogen input because the high nutrient input will enable a larger offshore primary production and a larger import or organic matter into the Wadden Sea. The relation between riverine Total Nitrogen (TN) load and the autumn concentrations of ammonium and nitrite was explored with a multiple regression analysis. Phytoplankton (as chlorophyll) was used as a co-variable because it can be expected that during autumns with a low light availability and low phytoplankton standing stocks (storms, cloudiness) ammonium and nitrite can reach higher values than during autumns with favorable light conditions and a high standing stock. Also temperature was taken in account as a factor enhancing the remineralisation rate. As a measure of the autumnal remineralisation intensity, we used mean values of the sum of ammonium plus nitrite during September, October and November.

Table 1. Results of a multiple regression with Total Nitrogen load via Rhine and Meuse as an independend variable, temperature and chlorophyll as co-variables and the mean ammonium plus nitrite concentrations in autumn in the western Dutch Wadden Sea as dependend variable

Variable	Beta	B	*p*
TN load	0.48	0.00003	0.012
Chl *a*	−0.42	−0.41	0.022
Temp	0.22	1.06	0.205

(N=19, p=0.0017, R^2=0.63, outlier: 1983).

Influence of riverine nitrogen loads on the annual nitrogen cycle in the Wadden Sea

Previous studies related Wadden Sea eutrophication with annual riverine nutrient loads (e.g. De Jonge, 1990). In the present study, no annual input data are used. Instead, the input data include the growing season and the preceding winter period. River input is based on the sum of the discharges measured at Maassluis and Haringvliet and mainly represents the discharges via Rhine and Meuse. Especially during periods of high river flow, discharge via the Haringvliet becomes important. In Figure 4, the Total Nitrogen load is presented as a month–year plot. The graph shows that the first large winter floods occur in December. On the basis of the conceptual model, it is expected that the total amount of nitrogen available before the onset of the phytoplankton determines the production potential in the coastal zone and the organic matter import into the Wadden Sea. For that reason, the December discharge was included in the total sum of nitrogen that may influence the annual primary production in the coastal zone. The monthly TN loads between December of the previous year and August were added and used to investigate the effect of TN load on the remineralisation intensity in autumn (September–November). This time series, also presented in Figure 4, shows large interannual variations up to a factor of two. Conspicuous features are the high loads during the mid-1980s, low loads during 1991–1993 and the high loads during 1994 and 1995.

Figure 5 presents the year–month plots of ammonium and nitrite in the Wadden Sea. The data set is based on monthly means. For comparison the riverine TN load is also shown. Both ammonium and nitrite

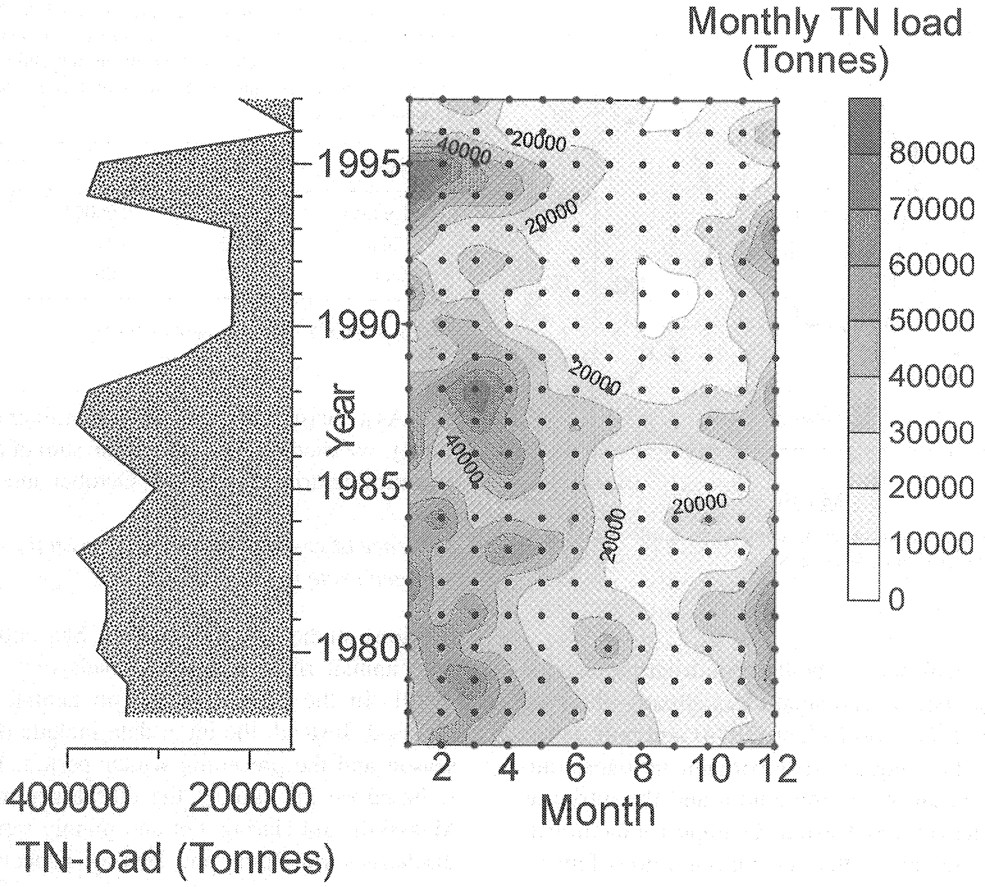

Figure 4. The monthly discharge of Total Nitrogen (TN) of the rivers Rhine and Meuse via Maassluis and Haringvliet (right panel). In the left panel, the time series of the total TN load from December (previous year) to August are shown.

have a clear seasonal cycle with low concentrations in summer and maximum concentrations during autumn. Highest autumn concentrations are found during the mid-1980s. Especially ammonium shows low values during the period of low TN discharges in the early 1990s. A multiple regression analysis was carried out with TN load as independent variable, temperature and chlorophyll as co-variables and the mean ammonium plus nitrite concentrations as the dependent variable. The analysis showed a significant correlation ($p<0.01$). Outliers can have a strong effect on the regression functions. One outlier (1983) could be identified having a distance from the model of 2.79 times the standard deviation of the other data and was rejected from the data set. The analysis again showed a significant positive correlation between riverine TN loads and autumnal ammonium plus nitrite values (Table 1). Phytoplankton (as chlorophyll) now had a significant negative effect on the autumnal ammonium plus nitrite levels presumably due to enhanced phytoplankton uptake. Low temperatures suppress the autumn maximum but the effect is not significant. Together the above factors explain 63% of the interannual variability of the autumnal ammonium plus nitrite levels. The analysis supports the hypothesis that in years with a high riverine nitrogen load more ammonium is released and more organic matter is remineralized than in dry years with a low riverine nitrogen load.

Figure 6 illustrates the effect of riverine nitrogen loads on the remineralisation levels in the Wadden Sea. In the five driest years (1991–1993, 1996, 1997), the riverine nitrogen load was 45% lower than during the five wettest years (1986–1988, 1994, 1995). The autumnal ammonium plus nitrit concentrations reflect this, being about 25% lower during dry years than during wet years. Apparently, the Wadden Sea eutrophication is not reduced proportional to the reduction of the riverine nitrogen load. Models show a similar reaction of the primary production in the Dutch coastal zone, where a reduction of the riverine nutrient

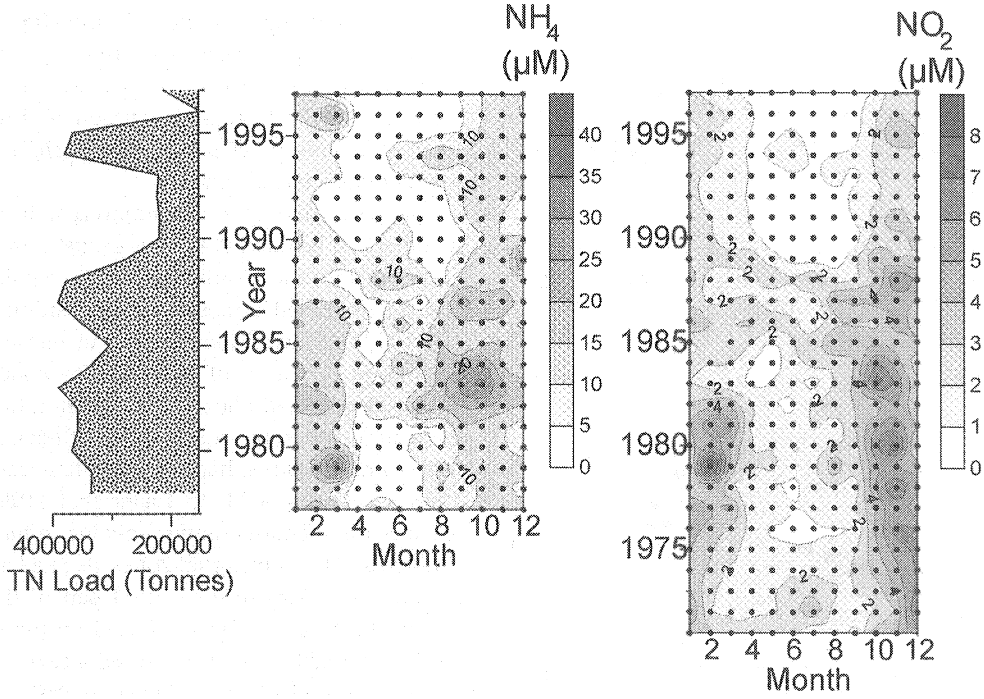

Figure 5. Month–Year Plot of ammonium and nitrite in the western Dutch Wadden Sea. For comparison the TN load (Rhine+Meuse) is shown on the left.

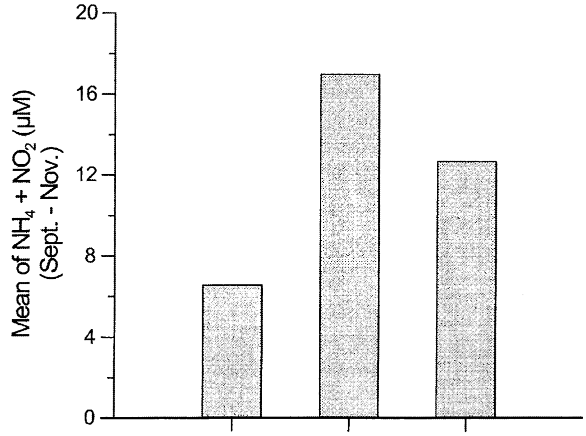

Figure 6. A comparison of the ammonium concentrations in autumn during the five wettest years (1986–1988, 1994, 1995) and during the five driest years (1991–1993, 1996, 1997) with 'historic' data from 1961 to 1962 (Postma, 1966).

loads of 50% yields a 10–30% reduction of the coastal primary production (ASMO, 1997; Lenhart, 1999). These results are in line with a proportional change of primary production in the North Sea and organic matter turnover in the Wadden Sea as formulated in the conceptual model.

A comparison with 'historic' data

The data analysis shows that at present the annual nitrogen cycle in the Wadden Sea responds immediately to changes in riverine nutrient load and a concomitant organic matter import. Compared to the first observations on the annual nitrogen cycle by Postma (1966) during 1960–1961 the present day values are two to three times higher (Fig. 6). In the following, it will be shown that this difference cannot be explained by changes in riverine nutrient input. Because of a lack of data the analysis based on the stations Maassluis and Haringvliet could not be extended to the early 1960s. To enable a comparison with riverine nitrogen loads, a time series of the river Rhine (Fig. 7a) measured near Lobith (Dutch–German border) was used. Dissolved Inorganic Nitrogen (DIN) data are available from 1952 onward, TN data from 1968 onward. A very significant correlation between Kjeldahl Nitrogen and ammonium (Fig. 7b) was used to reconstruct the pre-1968 TN load. The annual mean TN loads in 1960–1961 amounted to about 700–800 mol/s and were comparable to the TN loads observed during the 'dry' years 1991–1993 (about 700 mol/s). Despite comparable riverine nitrogen loads, the autumn remineralisation levels in the Wadden Sea during the

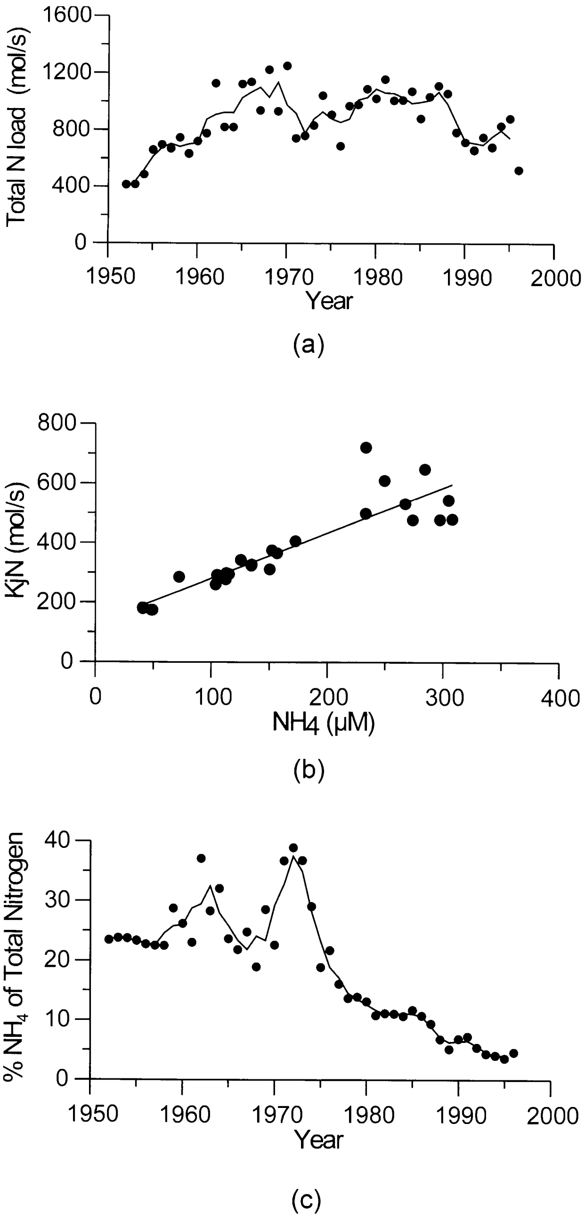

Figure 7. (a) Time series of Total Nitrogen in the river Rhine at Lobith (Dutch–German Border). Total nitrogen for the years 1952–1965 was not measured, but estimated on the basis of a very significant correlation between ammonium and total reduced (Kjeldahl) nitrogen shown in Figure 7b. (b) Correlation between ammonium and total reduced (Kjeldahl) nitrogen (1965–1992) in the river Rhine near Lobith (KjN=1.5∗NH4+129; r^2=0.80, n=26). (c) Relative contribution of ammonium to Total Nitrogen in the river Rhine (near Lobith).

dry years of the 1990s were about two times higher than during 1960–1961 and suggest a higher organic matter import from the North Sea. Two possibilities can be put forward to explain the higher organic matter import and higher remineralisation intensity in the Wadden Sea at present as compared to the early 1960s: (1) Increased nutrient concentrations at the saline end-member; and (2) decreased retention of nitrogen in the river Rhine and its estuary (resulting in an increased load into the coastal zone).

Increased nutrient concentrations at the saline end-member can be due to two factors, an increased nutrient input into the North Sea via the Strait of Dover and an increased atmospheric deposition. Laane et al. (1993) reviewed the changes in nutrient concentrations in the Strait of Dover. They concluded that winter nitrate (and phosphate) concentrations both in the central and in the coastal parts of the Channel and in the Strait of Dover had increased between 1960 and 1988 at salinities of 34–35. Laane et al. (1993) suggested that the increased anthropogenic influence in the English and French coastal zones also had an effect on the concentrations in the central part of the Channel and in the Strait of Dover. Based on the same data, De Jonge et al. (1993a) suggested a two- to threefold increase for both N and P since the 1960s. Apart from increased advection of nitrogen via the Strait of Dover, direct inputs via the atmosphere have increased. According to Gerlach (1989), the atmospheric deposition into the North Sea has increased by a factor of four since the early 1950s.

In conclusion, the off-shore nitrogen availability in the North Sea off the Wadden Sea might have doubled since the early 1960s. Assuming that nitrogen was the limiting nutrient during the 1960s, this would have doubled the primary production in the off-shore region. It is unclear how far off-shore the zone reaches from which the Wadden Sea derives its organic matter. The extent of this zone determines the relative contribution of river water and, therefore, the relative importance of marine and riverine nutrient enrichment. A simple calculation illustrates the importance of the increased nitrogen load through the Strait of Dover and atmospheric deposition on the off-shore primary production potential. Assuming that the imported organic matter is produced at a salinity of 30 (mean salinity at the entrance of the western Dutch Wadden Sea) and that Channel Water has a salinity of 35, a doubling of the nitrogen load via the Strait of Dover and by atmospheric deposition would increase the organic matter production by a factor of 1.85. For comparison, the riverine nitrogen load would have to increase by a factor of 6–7 to reach the same effect (cf. also De Jonge, 1997). Figure 7a shows that the riverine nitrogen loads increased by a factor of three at the most since the early 1950s. This suggests that the increased nitrogen input via the Strait of Dover and atmospheric deposition may be considered to be major factors to explain the increased organic matter import into the Wadden Sea.

In the above discussion on the influence of the Rhine/Meuse nitrogen loads on the eutrophication of the Wadden Sea, it was assumed that all riverine nitrogen reaches the coastal zone. This is not necessarily true. De Jonge et al. (2002) pointed out that a large fraction (20–50%) of the terrestrial nitrogen input is removed by denitrification. Billén et al. (1985) suggested that the implementation of secondary waste water treatment would change the oxygen status and the denitrification potential of rivers. This could increase the nitrogen loads into the coastal zone by a factor of up to two to three. The time series at Lobith indeed show that the nitrogen regime in the river Rhine has changed due to the implementations of waste water treatment plants by increasing the relative importance of nitrate (van Bennekom & Wetsteijn, 1990) and decreasing the relative proportion of ammonium and organic nitrogen (Fig. 7b, c). Which proportion of the riverine nitrogen load was lost in the Rhine/Meuse estuary during the during the 1960s due to denitrification is difficult to asses, but it was certainly less than the maximum factor of 3 proposed by Billén et al. (1985). The calculation presented above suggest that the river input had to be increased by a factor of about 6 to equal the effect of the doubled nitrogen input via the Channel. Clearly, this has not happened. Therefore, it is suggested that the increased remineralisation rates in the Dutch Wadden Sea as reflected by the increased ammonium plus nitrite concentrations in autumn between the 1960s and the 1980s/1990s are largely caused by an increased nitrogen flux into the North Sea through the Strait of Dover and via atmospheric deposition.

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Particulate inputs to Dublin Bay and to the South Lagoon, Bull Island

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Key words: particulates, nutrients, carbon, tidal exchange

Abstract

There are substantial riverine and sewage particulate inputs into Dublin Bay. The main river, the R. Liffey, accounts for almost 85% of the riverine input, which amounts to 37.1 t d^{-1} of SPM. The sewage input, which is discharged into the estuary, is slightly less at 35.3 t d^{-1}. The combined inputs deliver 17.4 t d^{-1}, 2.9 t $d^{-1}$and 1.2 t $d^{-1}$of particulate C, N and P, respectively, to the Bay as a whole. Although the particulate N load accounts for just 20% of the total N input, the retention of particulates within the system, and the subsequent remobilisation of N from the sediments is heavily implicated in the macroalgal blooms. A particulate mass balance of SPM, and of C, N and P for the South Lagoon showed no consistent pattern of import/export except that the fluxes of the individual elements reflected SPM load and varied with quantity rather than quality. The molar C:N ratio was around 6:1 although on occasion values approaching 3:1 were obtained, indicating very high quality and potentially labile SPM. The controlling factors in the budget appeared to be the water velocities and to a lesser extent, the SPM load, which surprisingly was no greater on a spring tide than on a neap. In this shallow, and primarily intertidal, system, wind- or wave-driven resuspension may exert more influence than predictable factors such as tidal range.

Introduction

Dublin Bay (Fig. 1), surrounded as it is on three sides by the Dublin conurbation which uses the Bay as the final destination for its sewage, nevertheless manages not only to maintain a largely normal fauna and flora (Walker & Rees, 1980; Wilson, 1982) but also to sustain a UN Biosphere reserve and bird populations of national and international importance (Jeffrey, 1977; ERU, 1992).

Opinions on the pollution status of the Bay have been equivocal (Wilson et al., 1990; Wilson & Elkaim, 1991; Wilson & Jeffrey, 1994), but seem to suggest that definite impacts can only be found with any degree of certainty in the inner areas of the Bull lagoons, the Tolka estuary and the Liffey estuary itself (Jeffrey et al., 1978, 1991; Wilson et al., 1986). However, both the littoral and the sublittoral experience eutrophication, in the form of excess macroalgal growths, suggesting some dysfunction in the system as a whole (ERU, 1992; Brennan et al., 1994).

Jeffrey et al. (1995) suggested two hypotheses, presented schematically below, to explain the production of the inshore green algae and the offshore brown alga *Ectocarpus* in Dublin Bay. Under the normal summer regime of light, temperature and salinity, nitrogen is the major limiting factor, since the irregular cessation of algal production in late summer (Jeffrey et al., 1995; Khan, 1998) points to exhaustion of a pool of readily mineralisable nitrogen in the sediment. This is similar to the pattern of labile or available nitrogen utilization observed in terrestrial soils. The key concept of both hypotheses is that nitrogen is transported to the production site mainly in particulate form. Intertidal production of green algae is then dependent on inorganic nitrogen supplied through mineralisation in the sediment (Jennings, 1996). In the subtidal areas, substantial amounts of macroalgae, mainly *Ectocarpus* were found to be anchored to the tubes of *Lanice conchilega*. In this hypothesis, particulate nitrogen is intercepted by the filter feeding activities of tubeworms which ingest the particulates, and subseqently re-released as excreted N or in the faeces.

Sedimentary geologists (Groen, 1967; Postma, 1967; Ward, 1981) developed the concept of ebb or flood dominated tidal flows and ecological interest in the transport of materials in estuaries resulted in the

Intertidal Green Algae	Benthic Algae
Organic Particulates	Flux of Organic Particulates
⇓	⇓
Sedimentation and Biogenic Uptake	Tubeworm Feeding
⇓	⇓
Pool of Organic N in Sediment	Catabolic Mineralization Excretion & Irrigation
⇓	⇓
Mineralization	Ammonium Release
⇓	⇓
Ammonium Release	Algal Assimilation
⇓	⇓
Algal Assimilation	Production and Reproduction
⇓	
Production and Reproduction	

Table 1. A summary of material (particulate and dissolved) transport studies in estuarine and lagoonal systems: + indicates import, − indicates export. (From Dame & Allen, 1996)

System	Partic.	Diss.	Reference
U.S.A.			
Bay of Fundy	−		Gordon & Cranford (1994)
Bedford Basin	−		Taguchi & Hargrave (1978)
Crommet Creek		+	Daly & Mathieson (1981)
Sippewissit	−	−	Valiela et al. (1978)
Flax Pond	+	−/+	Woodwell et al. (1979)
Canary Creek	−	−/+	Lotrich et al. (1978)
Gott's Marsh	−	−	Heinle & Flemer (1976)
Ware Creek	−	−/+	Axelrad (1974)
Carter Creek	−	−/+	Axelrad (1974)
Bly Creek	+	−	Dame et al. (1991)
North Inlet	−	−	Dame et al. (1986)
Cumberland Island	−/+	−/+	Childers (1994)
Sapelo Island	−	−	Chalmers et al. (1985)
Barataria Bay	−	−	Happ et al. (1977)
East Bay	−	−	Borey et al. (1983)
Biscayne Bay	−		Roman et al. (1983)
European			
Himmerfjard	+	−	Wilmot et al. (1985)
Ems-Dollard	+		Dankers et al. (1984)
Slufter	+	+	Asjes & Dankers (1994)
Stroodorpe	+		Wolff et al. (1979)
Zwin	+		Hemminga et al. (1992)
Colne Point	+	+	Abd. Aziz & Nedwell (1986)
Tollesbury	+	−	Boorman et al. (1994)
Mount St Michel	+	+	Boorman et al. (1994)
Mira	+		Boorman et al. (1994)

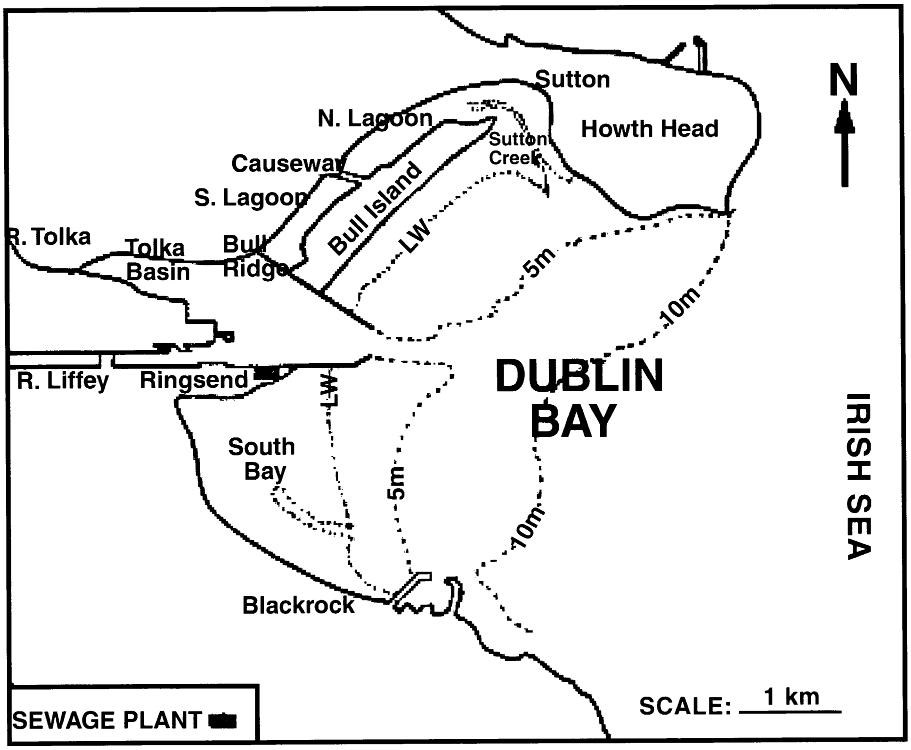

Figure 1 Dublin Bay, and the South Bull lagoon, showing major input sources (see also text).

proposal of the 'Outwelling Hypothesis' by Odum in 1968. This hypothesis states that marsh-estuarine ecosystems produce more organic material than can be utilised or stored within the system and that the excess material is exported to the coastal ocean where it supports near-coastal ocean productivity. Since this hypothesis was formulated, several investigations of material transport have been carried out and these have been summarised by Dame & Allen (1996). In European systems, there was usually a net import of particulate materials while in North American estuaries, export of material was normally observed (Table 1).

Two main approaches have been used in material transport studies. The indirect approach uses a mass balance budget of organic carbon provided by autotrophs (primary producers) and consumed by heterotrophs. If these production and consumption processes do not balance, then the transport of material to or from adjacent systems must be invoked (Hopkinson & Hoffman, 1984; Hopkinson, 1985, 1988). The direct flux technique involves measurements of SPM concentrations in the water column that are multiplied by water flow to provide estimates of material fluxes (Kjerve, 1975; Kjerve & Proehl, 1979). This technique has been used in SPM fluxes estimations in the Ems-Dollard estuary (Dankers et al., 1984), Mangere Inlet, New Zealand (Williamson et al., 1996), the German Bight (Puls et al., 1997) and others. A combined approach, with total system and major subsystems (such as saltmarsh) being observed simultaneously using a statistical design was applied by Dame et al. (1991) to the Bly Creek, South Carolina, U.S.A. This study demonstrated that different subsystems or combinations of subsystems influence or control the character of the various material fluxes.

The objective of this study was to quantify the influence of particulates on the Dublin Bay system, and to establish a balance for the particulates, and especially the nutrients, for the south Bull Island lagoon.

Materials and methods

The location of the study site, Dublin Bay is shown in Figure 1 along with the major sources of SPM to the Bay.

For the overall Bay study, the seven rivers and two canals which discharge to the Bay were monitored at approximately monthly intervals during 1993. Discharge rate data for the Liffey was obtained from the ESB and long term average flows as published in the WQMP study (ERU, 1992) were used for the Tolka and the Dodder.

Discharge rates for Ringsend sewage plant effluent were supplied by Dublin Corporation. The sewage effluent from the plant was monitored throughout 1993 at the rate of three samples per week for the first 6 months and four samples per month for the second half of the year. One litre samples of 24 h composite effluent were obtained from Dublin Corporation's Central Water Laboratory.

For the analyses, dissolved nutrients were defined as those in the filtrate passing through 0.45 μm filters, while particulates (SPM) were defined as those retained on a GF/C filter (Kramer et al., 1994).

Ammonium was determined with a Tecator dye (5000-0295) using a Tecator Flow Injection Analyzer, Model 5020 (detection limit was 10 μg l^{-1}). Nitrate (includes both nitrate and nitrite) was determined spectrophotometrically, following cadmium reduction/sulphanilamide reaction, using a Tecator Flow Injection Analyzer, Model 5020 (detection limit 10 μg l^{-1}). Phosphate (detection limit 1 μg l^{-1}) was measured by the ascorbic acid reduction method.

A 25-ml sub-sample was digested by the Kjeldahl method to determine kjeldahl nitrogen (KN) and total phosphorus (TP). The organic carbon and total nitrogen in the particulate component on the GF/C was determined on a LECO CNS-1000 analyser. The limit of detection was 0.02 mg g^{-1} for carbon and 0.02 mg g^{-1} for nitrogen.

Minor inputs, from drains and small streams were measured at high and low flow and atmospheric input estimated from Dublin City monitoring data.

For the Bull Island lagoon budget, the cross section under the bridge was surveyed and hourly current measurements taken for three tides at six stations across the channel. Replicate 1l samples were taken from the sub-surface water and the SPM collected on pre-treated GF/C filters. Dissolved and particulate nutrients were measured on four tides with samples taken as for the Bay study at 0.5 h intervals over the tidal cycle. Analysis for C, N and P followed the same procedures as for the Bay study (above).

Table 2. Discharge rates (m^3 x 10^3 d^{-1}) from Ringsend STW, and daily SPM (t d^{-1}) and particulate C, N and P load (kg d^{-1}) along with percentage contribution of the particulates to total nutrient loading

	Max.	Min.	Mean	% of total
Discharge	784	221	332	–
SPM (t d^{-1})	86.9	8.80	35.3	–
C (kg d^{-1})	35618	3255	14156	–
N (kg d^{-1})	5126	639	2206	30.0
P (kg d^{-1})	1496	127	595	35.4

Table 3. Discharge rates (m^3 x 10^3 d^{-1}) from the R. Liffey, and daily SPM (t d^{-1}) and particulate C, N and P load (kg d^{-1}) along with percentage contribution of the particulates to total nutrient loading

	Max.	Min.	Mean	% of total
Discharge (m^3 x 10^6 d^{-1})	5.81	0.63	1.94	–
SPM (t d^{-1})	202	5	37.1	–
C (kg d^{-1})	15373	683	3244	–
N (kg d^{-1})	2901	144	690	10.3
P (kg d^{-1})	2906	9	580	73.9

Results

Dublin Bay, as can be seen from Figure 1, is a very open system, and one which has considerable tidally-generated water movement. The mean tidal range is 2.75 m, which, applied over the extent of the Bay (33.75 km^2) gives an average of 9.3$\times$10^7 m^3 exchanged with every tide. This corresponds with a daily SPM and C load (at 47.5 mg l^{-1} and 1.83% (Wilson & Parkes, 1998)) respectively) of 8.3$\times$10^3 t d^{-1} and 151.9 kg d^{-1}, respectively. The matching figures for particulate (including organic) N and P, whose mean concentrations were 163 μg l^{-1} and 44 μg l^{-1} (Brennan et al., 1994), respectively, were 28.3 t N d^{-1} and 7.65 t P d^{-1}. These particulate inputs corresponded to 57% and 65% of the total N and P loadings from tidal exchange.

In addition to the tidal contribution, there are substantial inputs from Ringsend STW (Table 2) and from the rivers (Table 3) draining into Dublin Bay (Fig. 1).

The discharge of the river Liffey is at least an order of magnitude greater than those of the other rivers and accounts for almost 85% of the total an-

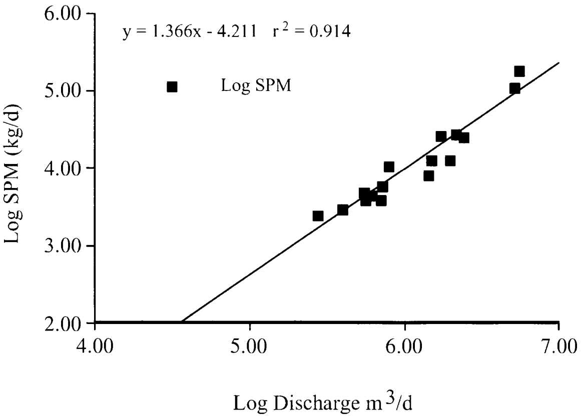

Figure 2. SPM load (kg d^{-1}) with discharge (m^3 d^{-1}) of the river Liffey.

Table 4. Particulate inputs (t d^{-1}) and percentage contribution to the overall Bay budget from river, STW and tidal exchange

	C	N	P
Tidal particulates	151.9	28.3	7.65
STW particulates	14.16	2.21	0.60
River particulates	3.24	0.69	0.58
All particulates	169.3	31.2	8.83
Particulate % of total	–	49.1	61.1

nual riverine input. The SPM input from the Liffey is governed largely by flow, such that the higher the flow, the greater the input (Fig. 2), although high flows are somewhat balanced by a slight increase in the inorganic percentage of the SPM. Nevertheless, the dominance of discharge is such that the C and nutrient inputs depend largely on the river flow.

The combined inputs from river and STW deliver a total of 72.4 t d^{-1} of SPM, of which 17.4 t d^{-1}, 2.9 t d^{-1} and 1.2 t d^{-1} are particulate C, N and P respectively. In addition, there are other minor inputs. Atmospheric N inputs are estimated at 2% of the total, and there are a number of small streams whose input may be locally important, especially in times of high flow, and the same may be said of groundwater. However, these are negligible on the scale of the whole of the Bay, and the particulate inputs are summarised in Table 4.

In the case of the south lagoon (Fig. 1), it was possible to get a more accurate estimate of the particulate input through simultaneous measurement of current speed and particulate load passing under the bridge over a tidal cycle. Figure 3 shows the data from 3 tidal cycles; (A) tidal range 2.53 m; (B), tidal range 2.11 m; and (C) tidal range 3.13 m.

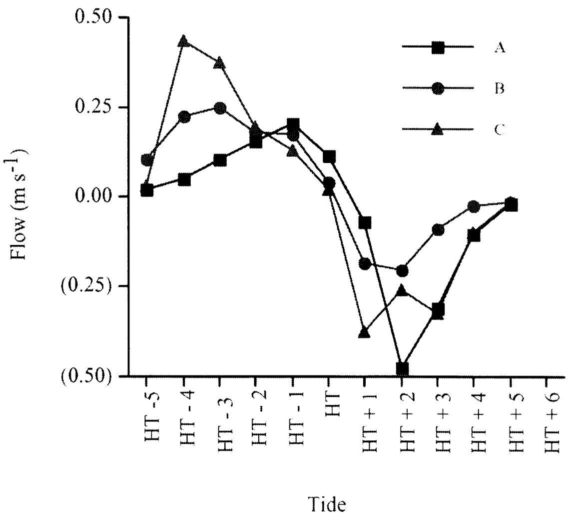

Figure 3. Velocity (m s^{-1}) of water flow under Bull Island bridge.

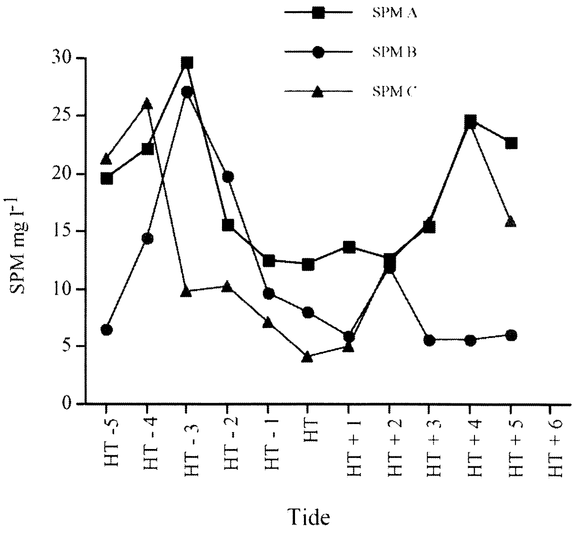

Figure 4. SPM loads (mg l^{-1}) under Bull Island bridge.

The tidal pattern of the flows can be clearly seen (Fig. 3) with a quasi-symmetry around HT. However, there were considerable difference between the tides, with for example the flows of tide A being much stronger on the flood than on the ebb.

The SPM loadings varied over the tidal cycles (Fig. 4) but a consistent pattern was evident, with the lowest loads around high- and low-water when flow rates were at their lowest.

However, a note of caution has to be entered here, as the correlation between SPM load and flow was nowhere near as clear as with the river data shown

Table 5. Hourly SPM transport (kg), total input (t) and output (t) and overall balance for each of the three tides

State of tide	Tide A	Tide B	Tide C
LW	0	0	0
HW −5	+47.2	+287.7	+62.9
HW −4	+301.7	+1167.1	+1972.6
HW −3	+2225.5	+2848.8	+9500.7
HW −2	+2401.6	+1137.0	+2192.8
HW −1	+3019.6	+1263.7	+1703.4
HW	+1873.5	+360.0	+255.8
HW +1	−1260.8	−865.5	−2220.0
HW +2	−6278.3	−851.0	−1551.5
HW +3	−3765.9	−909.4	−2345.8
HW +4	−1779.8	−393.0	−366.8
HW +5	−93.9	−82.7	−55.3
Input (+, t)	9.87	7.06	16.29
Output (−, t)	13.18	3.10	6.54
Net Balance (t)	−3.31	+3.96	+9.75

in Figure 2. In fact, there was almost no correlation (r=0.016) between absolute flow rate and SPM concentration, but a slightly stronger, though still not significant, correlation (r=0.22) was obtained by differentiating inward and outward flows. This is demonstrated in Figure 4 where the SPM loads on the flood (inward) are higher than those on the ebb (outward).

The third factor in the flux calculation is the depth and width of the channel under the bridge. This varied from a small pool of static water at low tide of just over 35 m^2 in cross section to a maximum of 560 m^2 at high water on the highest tide (tide C). The hourly transports and the net balance are shown in Table 5.

The final balances show no consistency in the SPM fluxes. Tide B, the smallest tide, and tide C, the largest tide were net importers, but tide A, which was intermediate, was a net exporter. However, the amounts transported by tides A and C were broadly similar, albeit in different directions, and both were markedly higher than tide B.

In terms of particulate C and nutrient fluxes, the balances were strongly influenced by the amounts of SPM transported. Nevertheless, there were marked differences in C and N content both among tides and over the tidal cycle (Fig. 5a, b).

Tide B, the smallest tide had the highest SPM C content, and also a marked difference between the inflow which was much lower in SPM C than the outflow. This quality difference was reversed for SPM N, for which tide B showed much higher inflow levels than outflow. These differences were not so marked for the other two tides, nor were any marked patterns seen in SPM P, for which only tides A and B were measured. SPM quality did not correlate with flow rate in or out, for either C (r=0.10), N (r=0.03) or P (r=0.24). There was no overall correlation of SPM N with SPM C (r=0.20) but SPM P was significantly (r=−0.48, p=0.02) negatively correlated with C.

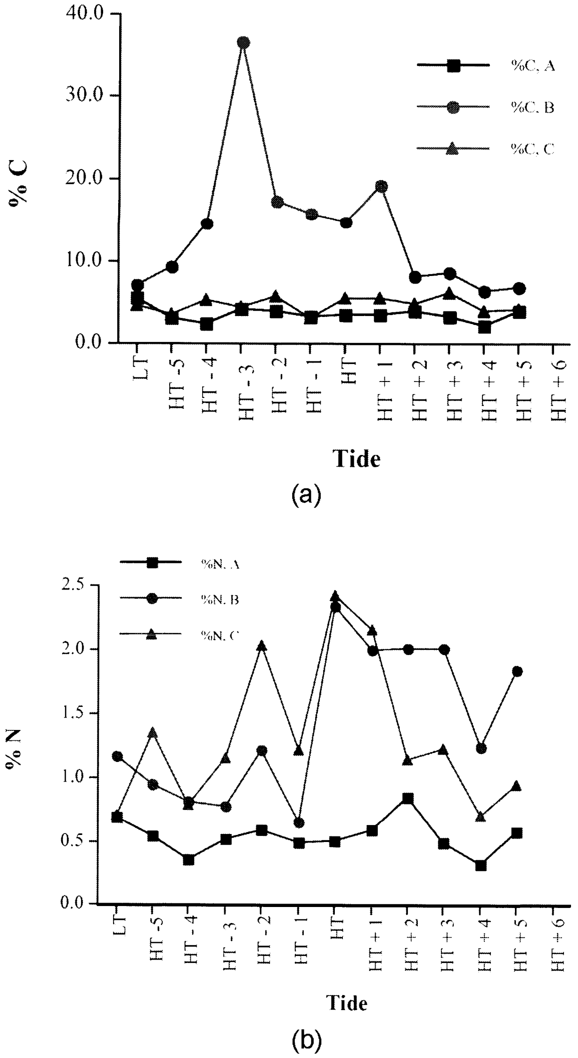

Figure 5. (a) SPM% C content and (b) SPM % N content over the tidal cycle for tides A, B and C.

The overall balances of C, N and P are shown in Table 6. Despite the lack of direct correlation with SPM, it is evident that the transport pattern of C and of the nutrients follows that of the SPM. A net input of

Table 6. C, N and P: total input (kg) and output (kg) and overall balance (kg) per tide for each of the three tides

	Tide A C	Tide B C	Tide C C	Tide A N	Tide B N	Tide C N	Tide A P	Tide B P
LW	0	0	0	0	0	0	0	0
HW −5	+1.47	+26.99	+3.01	+0.258	+2.704	+0.438	+0.002	+0.506
HW −4	+7.48	+170.06	+71.78	+1.086	+9.512	+26.591	+0.527	+1.324
HW −3	+95.72	+1046.7	+497.45	+11.42	+22.021	+75.245	+0.524	+3.082
HW −2	+98.13	+197.62	+97.23	+14.15	+13.792	+25.174	+1.085	+0.463
HW −1	+103.88	+201.07	+99.26	+14.95	+8.201	+34.511	+6.317	+0.388
HW	+68.10	+53.14	+7.86	+9.48	+8.402	+3.108	+5.142	+0.000
HW +1	−45.03	−166.71	−125.96	−7.40	−17.250	−53.724	−0.817	−0.051
HW +2	−258.60	−68.97	−85.29	−53.37	−17.045	−33.357	−6.253	−0.321
HW +3	−123.26	−78.201	−112.60	−18.83	−18.225	−26.648	−6.059	−0.488
HW +4	−40.83	−25.13	−23.12	−5.695	−4.881	−4.515	−2.001	−0.019
HW +5	−3.86	−5.726	−2.230	−0.542	−1.522	−0.388	−0.018	−0.663
Input (+)	374.78	1695.6	776.59	51.344	64.632	165.07	13.597	5.763
Output (-)	471.58	344.7	349.20	85.838	58.92	118.63	15.148	1.542
Net Balance (kg)	−96.80	+1350.9	+427.39	−34.494	+5.709	+46.44	−1.551	+4.221

SPM (Table 5) is reflected in a net input of C and N, although for tide B, there was a net outflow of particulate P.

The hourly flux data in Tables 5 and 6 show, in additional to the tidal pattern, that a single calculated hourly flux can exert a prime influence on not only the magnitude, but also the direction of the overall balance – see for example tide A (HW+2), and tide B (HW-3).

The quality of the SPM varied also. Figure 6 shows the molar C:N ratio in SPM from tides A, B and C (as above) along with the 1993 data (tides D, E, F, G with tidal ranges 3.59 m, 2.32 m, 3.81 m and 2.60 m, respectively). The most striking difference (Fig. 6) was between tide B in which the C:N ratio reached 55.45 and the others, in which the C:N ratio mostly ranged between 5 and 10. The C:N ratios of the two winter neap tides (A, B) were higher than the summer tides (D–G) but the winter spring (C) was mostly lower and in addition displayed a marked tidal cycle. There is some suggestion in Figure 6 that the outgoing SPM had lower C:N ratios than the incoming, particularly so in tide B, but this was rather a weak trend, and there was no overall significant correlation between the tidal range and C:N ratio of the SPM.

The balance between dissolved and particulate nutrients with tidal state is shown in Figure 7. The ratio for P (Fig. 7b) showed little consistent pattern over

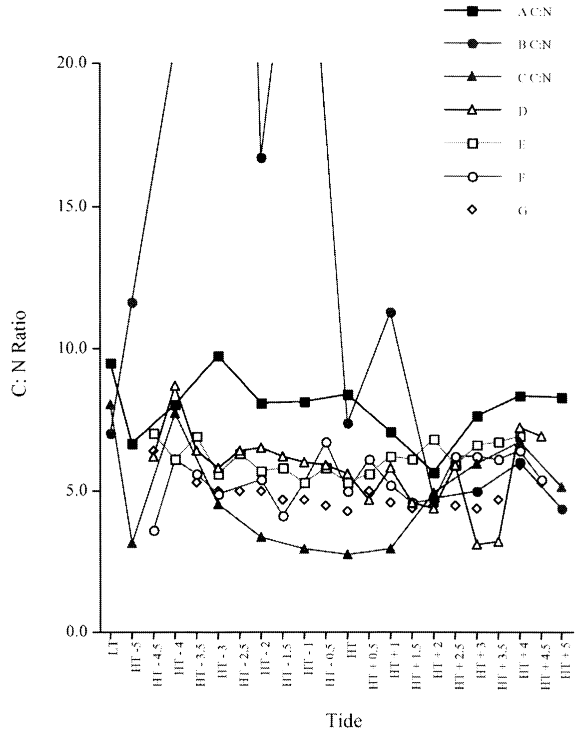

Figure 6. C:N ratios of tides A–G with tidal stage: see also text for explanation.

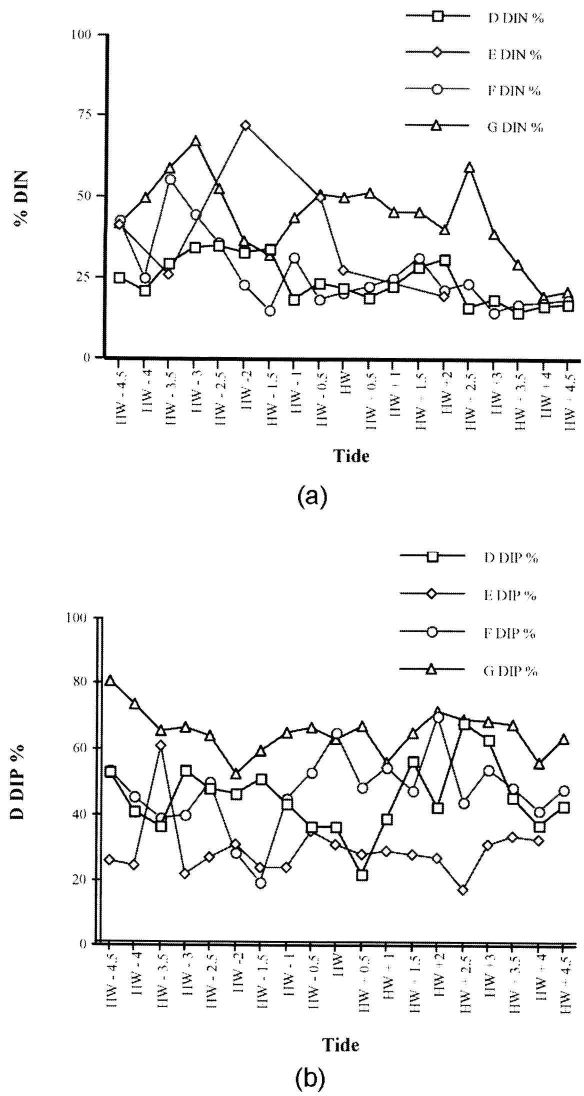

Figure 7. Percentage of nutrients in dissolved form over the tidal cycles for tides D–G: (a) DIN/Total N; (b) DIP/Total P.

the tidal cycle and the DIP varied from a minimum of around 20% to a maximum of 80%. The tide with the lowest tidal range (Tide E, 2.39 m) had the lowest % DIP over the whole of the cycle, but the next lowest range tide (Tide G, 2.60 m) had the highest % DIP. Nitrogen showed more of a pattern, with DIN ratios generally lower on the ebb than on the flood. This was actually most obvious for Tide E, although not evident on Figure 7a, in that both NO_3^- and NH_4^+ were near or below detection limit for many of the measurement intervals on the ebb. Figure 7a also emphasises the role of particulates in N transport, with most of the samples <50% DIN and some <25%.

Using the SPM load and quality data of Tides D–G (Brennan et al., 1994) did not change the direction of import or export, although the amounts naturally altered.

Discussion

The importance of particulates in the Dublin Bay system reflects not only the shallow and dynamic nature of the Bay, but also the considerable inputs from both sewage and river. Wilson & Parkes (1998) highlighted the influence of the particulate input in the overall energy budget of the Bay and suggested that it was in fact the driving force behind both the productivity and the structuring of the food web.

The particulates do play a major role in the delivery of C, N and P to the Bay system. From Wilson & Parkes' (1998) calculations, primary productivity in the Bay is equivalent to some 4.36 t C d^{-1}, or about 25% of that delivered by river and STP. The land-derived inputs, at around 10% of the total, are in turn dwarfed by the marine contribution from water exchange with the Irish Sea system over the seaward boundary. A similar picture exists for N and for P, although for the latter, the riverine and STP contribution is proportionately more at just over 15%.

The influence of the particulates on the functioning of the Bay is a consequence of their behaviour and fate, and the hydrodynamic model of the Bay (Brennan et al., 1994) suggested that the offshore area of high deposition of particulates coincided with the area of highest worm tube and hence *Ectocarpus* density, thus providing indirect support for the macroalgal growth hypothesis (Jeffrey et al., 1995).

More direct evidence is available in the case of the intertidal macroalgal blooms in the Bull Island lagoons. Various studies (Brennan et al., 1994; Jeffrey et al., 1995; Jennings, 1996; Khan, 1998) have investigated nutrient balance and nutrient flux in algae and sediments and shown that maximum growth in the algae coincides with maximum ammonium release from the sediments. The nutrient-laden sediments accumulate in the lagoon over the winter and the nutrients are remineralised by microbial (and invertebrate) metabolic activity with the warmer summer temperatures. That the dissolved nutrient supply cannot meet peak demand is shown by the fact that in the north Bull lagoon, which does not have the direct connection to the River Liffey or the STP discharge, N levels in algal tissues actually fall below the growth threshold of 2% (Khan, 1998).

However, the budgets presented here, while showing substantial nutrient transport in particulate form, do not fully support the hypothesis of inward particulate nutrient transport. While two of the three tides studied showed import, the third showed a net export, and this pattern was the same even with different SPM data. The principle controlling factor appears to be the water flow, rather than SPM load or particulate quality.

It is suggested that the importance of the particulates in the system lies not so much in the amounts of C, N and P that are delivered, but their fate. In comparison with other systems, the actual levels of SPM in the waters of Dublin Bay are rather low, reflecting the predominantly sandy nature of the habitats. However, the focussed inputs from river, and more importantly STP, appear to fuel localised eutrophication – the average C:N ratio of the SPM sampled under Bull bridge was close to 8:1 which is very close to that of the SPM from the STP.

Unlike the river, there appeared little correlation between SPM loads and water flow in and out of the lagoon. The SPM load may thus reflect stochastic factors such as wind strength, and even direction, especially given that both inside and immediately outside the lagoon are stretches of tidal flats.

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A model for predicting the temporal evolution of dissolved oxygen concentration in shallow estuaries

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Key words: depth averaged transport, dissolved oxygen, modelling, shallow estuaries, Urdaibai estuary

Abstract

The design of sewage discharge systems in estuaries needs to consider the dissolved oxygen concentration among other water quality indicators. Due to the great number of factors affecting the dissolved oxygen, the prediction of the temporal evolution of this element requires the use of mathematical tools. In the case of shallow estuaries with extensive intertidal zones, the complexity of this task increases since the water domain varies continuously. This work describes a numerical model which solves the vertical integrated transport equation including the effect of extensive tidal flats. The model makes use of the tidal velocities computed by a previous run of a hydrodynamic model. This procedure allows the dissolved oxygen modelling to be performed using a larger time step than that used for the velocity field calculation. The numerical scheme developed for the model guarantees that even in the drying and wetting of intertidal areas the water column dissolved oxygen concentration is not modified by numerical errors. The representations of the processes included in the model (advection, dispersion and reaction) have been validated successfully in several theoretical cases. An application to the Urdaibai Estuary, a shallow estuary in the Basque Country, Northern Spain, is also presented.

Introduction

Nowadays estuaries play an important role in social and economic activities. Some of the most populated and famous cities of today are located at estuarine banks. In recent years, the continuous degradation of estuarine ecosystems and the search for an equilibrium between sustainable development and environment has powered the achievement of water quality studies and sewage system works all over the world. Dissolved oxygen concentration has traditionally been used as a variable of water quality. This choice is based on the relationship observed between its concentration decrease and the appearance in the water column of non-desirable effects. The great number of factors influencing dissolved oxygen levels has led to consider the use of complex mathematical tools for studying and predicting their evolution.

Shallow estuaries are identified by the presence of wide tidal flats which yield an important variability in the water domain. This feature increases the difficulty of dissolved oxygen modelling, constituting a restriction in the application of many water quality models that have been successfully validated in environments in which water domain changes have no appreciable effects (deep estuaries and bays, coastal areas, closed seas).

In these shallow estuaries characterized by a two dimensional flow and important water depth changes in cross section to main flow, the temporal dissolved oxygen evolution analysis needs to solve the transport equation for each substance. Its most general expression is given by:

$$\frac{\partial c}{\partial t} + u_i \frac{\partial c}{\partial x_i} = \frac{\partial}{\partial x_i}\left(D_{ij} \frac{\partial c}{\partial x_j}\right) + R, \qquad (1)$$

where c is the concentration; u_i is the velocity components; D_{ij} is the diffusion coefficients matrix; R is the sources and sinks. Thus, it is possible to evaluate the influence of the overall process on the concentration balance.

In those cases in which it is possible to assume that estuarine water is well mixed vertically, Equation

(1) can be integrated over the depth, resulting in the following expression:

$$\frac{\partial(HC)}{\partial t}+\frac{\partial(HU_iC)}{\partial x_i}=\frac{\partial}{\partial x_i}\left(H\varepsilon_{ij}\frac{\partial C}{\partial x_j}\right)+RH, \tag{2}$$

where C is the depth-averaged concentration; U_i is the depth-averaged velocity components; H is the water depth and ε_{ij} is the dispersion coefficients matrix that includes the effect of the velocity profile depth averaging.

Sometimes the above equation is expressed using the water mass conservation principle (or continuity of water mass) as follows:

$$\frac{\partial C}{\partial t}+U_i\frac{\partial C}{\partial x_i}=\frac{1}{H}\frac{\partial}{\partial x_i}\left(H\varepsilon_{ij}\frac{\partial C}{\partial x_j}\right)+R. \tag{3}$$

This expression is a non-conservative form of the advection-diffusion equation as velocity gradients in the coordinate directions are neglected. This equation is exact only if water depth is constant and therefore currents are uniform, so in shallow areas by using Equation (3) the principle of substance mass conservation is not satisfied and also it could cause numerical instability (as water depth approaches zero and depth gradients increase).

Hence, numerical solving of vertically averaged transport processes in shallow estuaries needs to consider the transport equation in its more general expression as shown in Equation (2).

Model description

The model described in this work allows Equation (2) to be solved by using an explicit finite difference scheme based on the split operator approach in which the advection and diffusion processes are computed independently for each time step. This approach makes it easy to use different numerical methods for solving each process (Komatsu et al., 1997). Hence, advective transport is computed by an upwind scheme while diffusion is described by a centred scheme. The choice of an explicit scheme to solve the depth averaged transport equation places restrictions on the time step needed for guaranteeing numerical stability (Koutitas, 1988) resulting in a maximum value for the time step in the order of a few minutes.

In a forward step, the water quality variable concentration change caused by reaction processes is computed. This calculation would make it necessary to solve a set of differential equations (one equation per water quality variable) for each time step. To reduce the computational cost required by this method in the model, it has been considered that, in the same time step, only the concentration of the actual study variable is modified, holding constant the concentration level of the others. This hypothesis is valid only when the reaction process rates are slow, as occurs for short time steps, and leads to single expressions that can be solved analytically in the model. Such expressions have the following form:

$$\frac{\mathrm{d}C}{\mathrm{d}t}=A_1C+A_2, \tag{4}$$

where A_1 represents a set of terms proportional to the study variable concentration; A_2 represents a set of terms that includes other processes not depending on the study variable concentration.

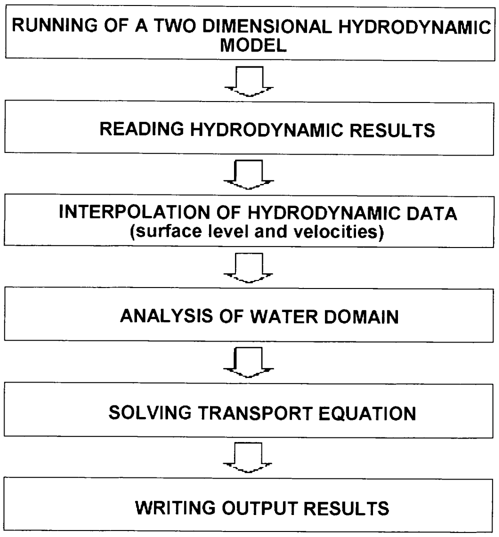

Figure 1. General model working scheme.

The model reproduces estuarine water movements during a tidal cycle using shorts time steps (in the order of minutes) from hydrodynamic model results (velocity components, water surface level) obtained by a previous running of a two dimensional hydrodynamic model. This working scheme (Fig. 1) allows a larger time step to be selected for the dissolved oxygen modelling than that used for the velocity field calculation. But in the case of shallow estuaries, this methodology has the disadvantage that during hydrodynamic data interpolation inside the water quality model there is an information loss about the real in-

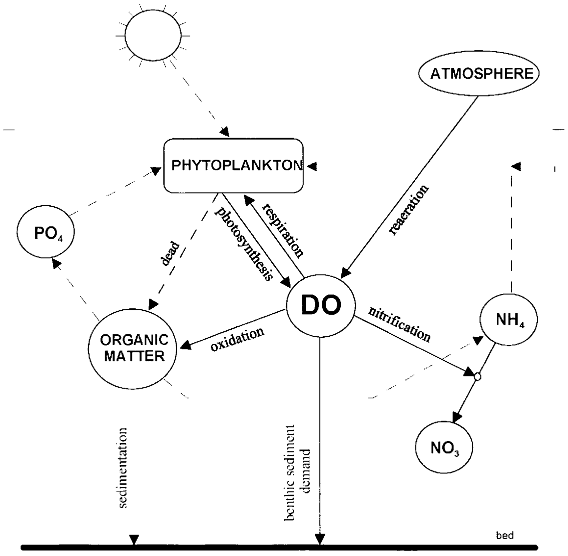

Figure 2. Processes included in the model as main mechanisms responsible for dissolved oxygen changes in estuarine water column and variables used for concentration balance.

stant in which wetting and drying of the tidal flats occurs.

To avoid this problem caused by using hydrodynamic results stored separately in time, a subroutine has been included in the model that allows the characteristics of the water surface level to be characterized in the whole estuarine domain between two consecutive recordings. In this way the exact instant in which shallow areas get wet or dry is accurately computed.

Variables and processes included in the model as main mechanisms responsible for dissolved oxygen changes in the water column are shown in Figure 2.

About the representation of these processes it is interesting to point out that:

1. Oxygen exchange between atmosphere and the water column is considered proportional to the dissolved oxygen deficit. Also the additional effect due to wind induced internal turbulence is accounted for.
2. Organic matter oxidation, nitrification and denitrification processes are computed using a first order kinetic considering the influence of the dissolved oxygen levels.
3. The phytoplankton growth rate is expressed as the difference between gross production and mortality plus sedimentation.
4. The gross growth rate is considered to be dependent on three variables (temperature, solar radiation, nutrients) assuming that these effects are multiplicative (Thomann & Mueller, 1987). Temperature effect is described by an arrhenius expression (Eppley, 1972). Solar radiation effect is defined by the depth averaged photoinhibition curve of Steele, 1962. Nutrient effect is described as a function of inorganic nutrient availability using a Monod growth kinetics.
5. The phytoplankton death rate includes the phenomena of endogenous respiration, zooplankton grazing and bacterial mortality.

Model test

The discretization used to compute the different processes included in the model has been verified in several theoretical situations. These situations have been used to analyse the numerical implementation of the three processes included in the transport equation: advection, diffusion and reaction. Tests have also been made on the procedure adopted to simulate these processes in shallow areas. An application to the Urdaibai Estuary, a shallow water body in the Bay of Biscay (Northern Spain) is also shown in this paper.

Advective transport

In general, the upwind scheme has a numerical diffusion depending on the spatial discretization selected for the water domain and the time step used in the computation. This dependence is expressed by the Courant number, defined as follows:

$$Cr = u\frac{\Delta t}{\Delta l}, \tag{5}$$

where Cr is the Courant number; u is the advective velocity; Δt is the time step; Δl is the characteristic spatial discretization length. For those cases in which the Courant number approaches to one, numerical diffusion is negligible.

In most estuaries, the horizontal tidal dispersion (Zison et al., 1978; Fischer et al., 1979) is greater than the diffusion caused by the numerical scheme.

Diffusion processes

The results given by the centred numerical scheme used to solve dispersion processes have been verified in a set of theoretical scenarios. In some of them the bed was assumed to be flat while others had depth

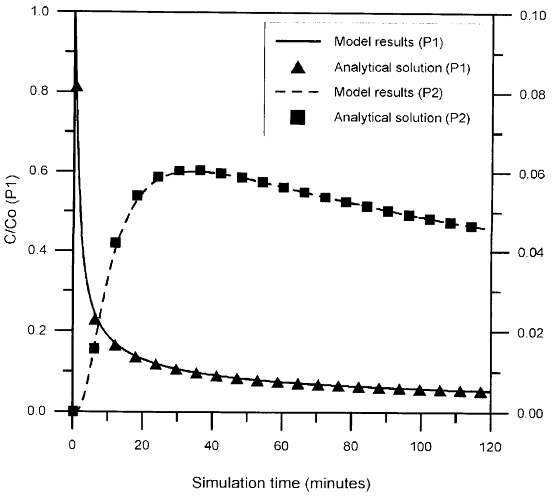

Figure 3. Comparison between model results and analytical solution in the case of two dimensional dispersion over flat bed.

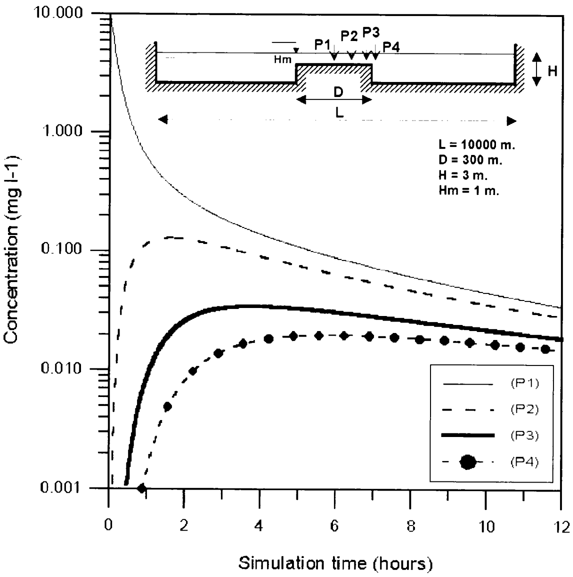

Figure 4. Temporal evolution concentration curves for two dimensional dispersion over variable bed.

gradients. In the first case, the model results were compared with their corresponding analytical solutions. An example of these situations is given below. For this analysis, a single concentration pulse (C_0) was initially introduced at the centre point of a squared pond, and the temporal evolution of the discharged concentration was studied. Figure 3 shows a comparison between the data obtained with the model and the analytical solution results at the discharge point (P1) and a point located 50 m away (P2).

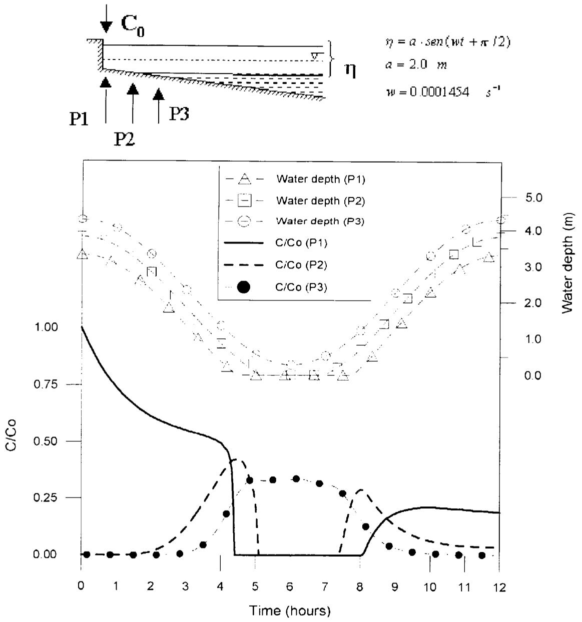

Figure 5. Temporal evolution of concentration and water depth as a result of model application to an estuary with lineal depth variation in which water surface level experiments a semidiurnal sinusoidal movement.

For the second theoretical cases group, it was verified that depth gradients did not cause numerical instability, as shown in Figure 4.

Wetting and drying phenomena

To check the accuracy of the scheme adopted for solving wetting and drying processes in shallow areas, several situations have been proposed. As an example, Figure 5 shows the results obtained from model application to a lineal estuary with water surface level oscillations. A concentration pulse, C_0, was initially discharged into the shallowest part of the estuary at high tide. Results indicates that even close to the drying of shallow cells, new maxima concentration peaks are not observed. Also substance mass conservation is fulfilled.

Reactive processes

This paper presents one of the numerous scenarios proposed to test the methodology used for solving reaction processes. This situation studies the influence of phytoplankton biomass evolution on dissolved oxygen levels and their dependence on nutrient availability. The hypothesis that a nutrient point source (ammonium, nitrate and phosphate) was initially in-

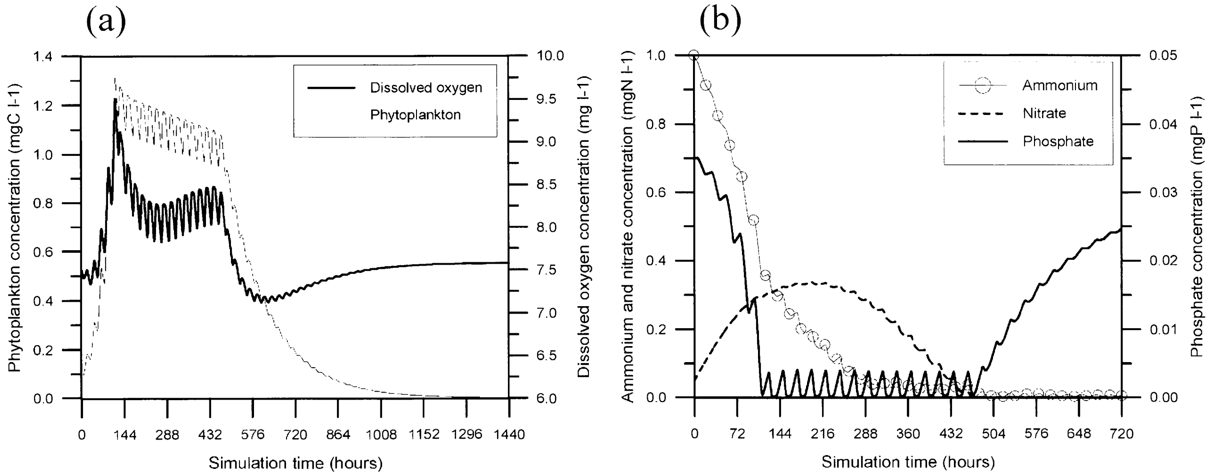

Figure 6. Temporal concentration evolution curves obtained from proposed mathematical model: (a) dissolved oxygen and phytoplankton biomass; (b) ammonium, nitrate and phosphate.

troduced in the water column was adopted. Also, the biochemical oxygen demand and the sediment oxygen demand were considered negligible. Results provided by model running (Fig. 6) reflect that, in a first stage, a huge phytoplankton growth took place due to a great nutrient availability, especially of ammonium. This inorganic compound suffered at the same time a nitrification process accentuated by high dissolved oxygen levels. After this stage, there is a decrease in phytoplankton biomass caused by an available nutrient reduction, which became more pronounced when the nutrients were almost consumed. At the end of the simulation, the dissolved oxygen concentration reached the saturation value. Also, there was no presence of ammonium because it was completely oxidized to nitrate.

Model application to the Urdaibai Estuary

The Urdaibai Estuary is a mesotidal temperate estuary located in the Bay of Biscay, Northern Spain (Fig. 7). An important geomorphological feature is its shallowness (on the average 3 m deep in the main channel). The estuary is well-mixed to partially-mixed depending on river discharge and tidal range. Estuarine waters receive the discharge of a domestic sewage treatment plant effluent in the upper estuary (near Gernika) that introduces a considerable amount of nutrients (mainly ammonia and phosphate) to the estuary causing eutrophication conditions (Ruiz et al., 1994). The geometry and bathymetry of the estuary was resolved using rectangular finite-difference cells of dimension 30×100 m. The resulting model grid contained 190×145 cells.

The hydrodynamic model was calibrated and validated with the data (water level and velocity) measured between January 1998 and February 1998 in nine stations distributed along the estuary (CHN, 1999). The results obtained showed that the hydraulic model reproduced water movement satisfactory in the whole estuary.

To test model applicability to the estuary, samples at high tide were taken on four consecutive days (24, 25, 26 and 27 May 1999) at the stations marked in Figure 7. Averaged river discharge in this sampling period was less than 0.5 m^3 s^{-1}, and the tidal range varied between 2.0 m and 2.7 m.

As shown in Figure 8, the computational results obtained agree with measured data in estuarine waters. Thus, the model could be used as a tool for computing algal growth dynamics and other water quality processes affecting dissolved oxygen concentration in shallow water bodies.

Conclusions

A two dimensional mathematical model for computing dissolved oxygen concentration changes in estuaries, considering the water domain variability related to wetting and drying phenomena of shallowest areas, has been developed. Transport processes are solved from the depth averaged transport equation in its more general expression. An explicit finite difference

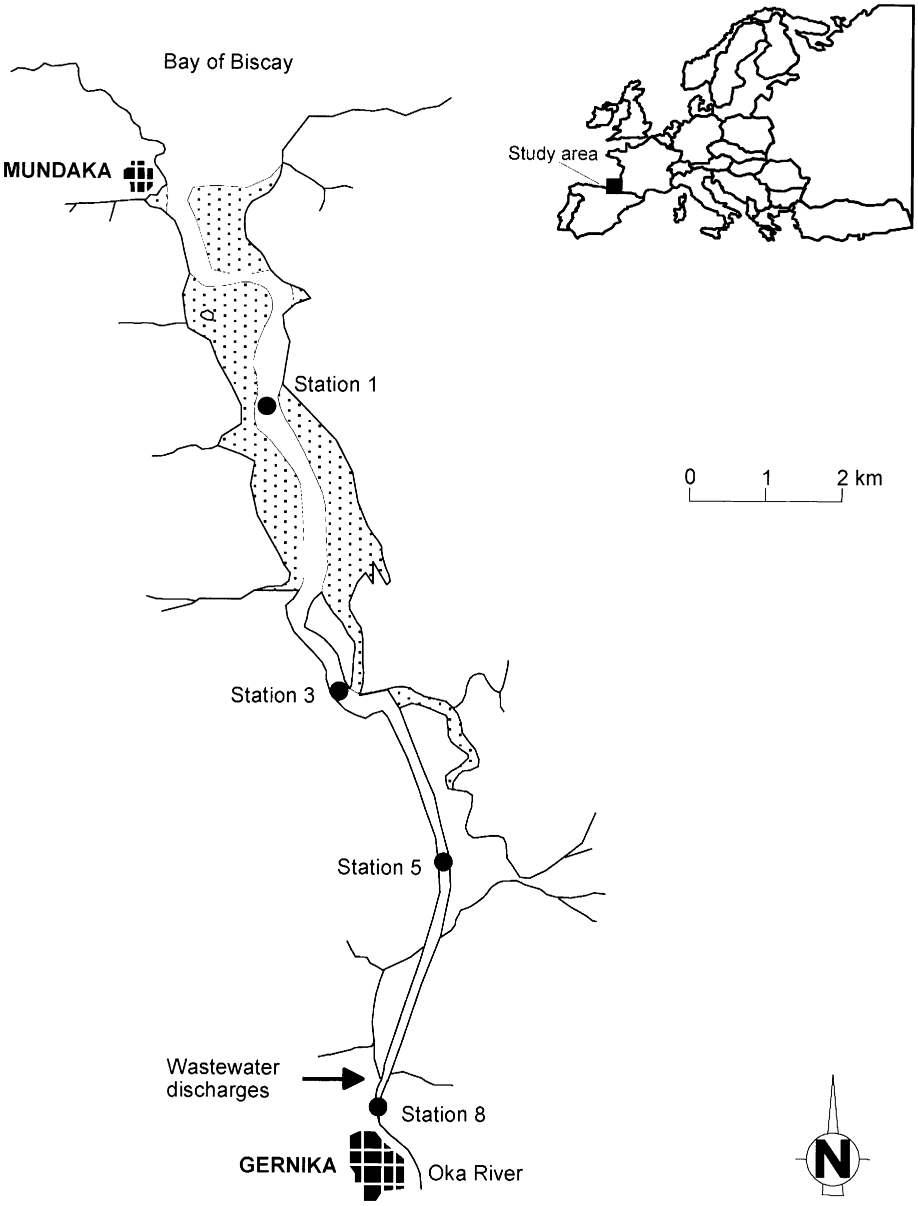

Figure 7. Study area and sampling stations(●).

scheme based on the split operator approach has been used to evaluate mass transport. The application of the model to different proposed scenarios shows that the methodology used to solve the transport equation for each water quality variable led to accurate results free of numerical instability. Results obtained from its application to a shallow estuary with a complex bathymetry (Urdaibai Estuary, Northern Spain) agree with data measurements.

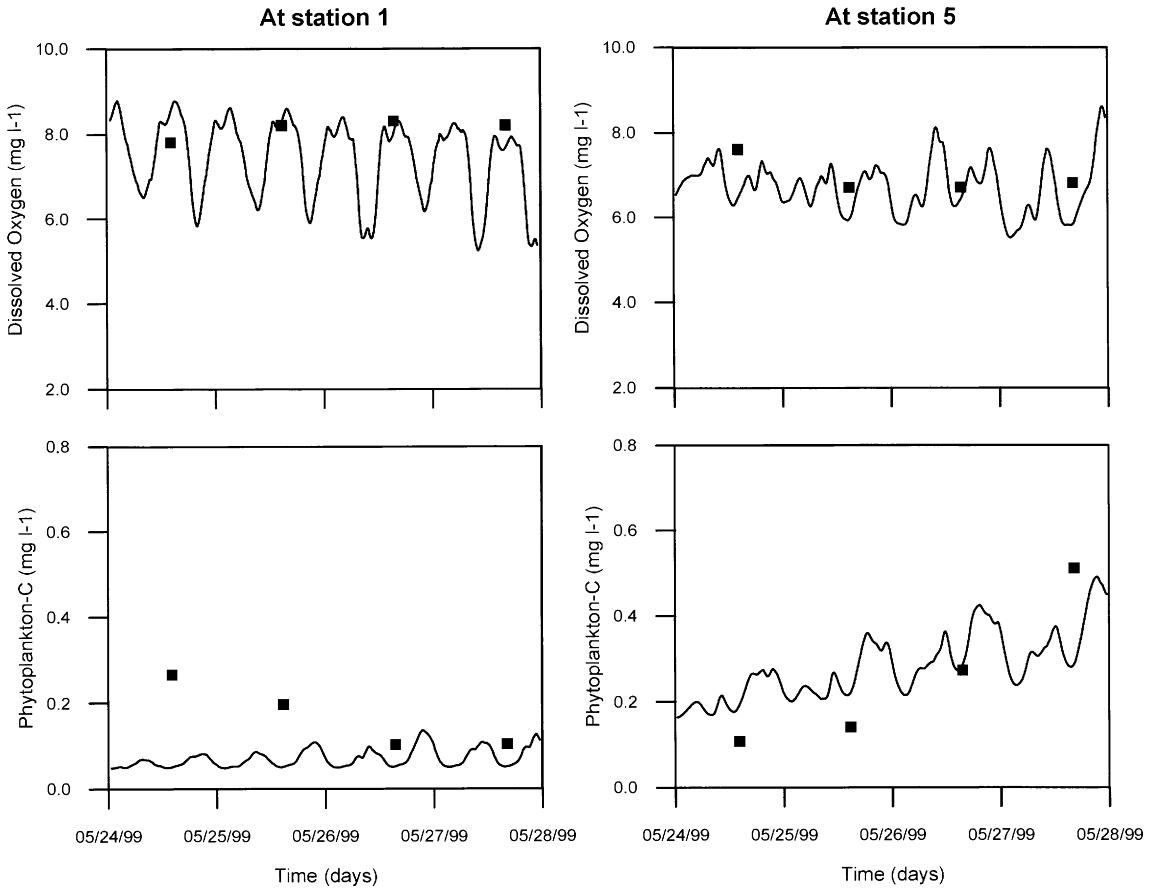

Figure 8. Dissolved oxygen and phytoplankton-C computed (—) and measured (■) at stations 1 and 5 in Urdaibai Estuary (Northern Spain) from 24 May to 27 May 1999. Depth averaged values.

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Modelling the faecal coliform concentrations in the Bilbao estuary

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Key words: faecal coliforms, modelling, bathing waters, sewerage scheme

Abstract

Since 1984, an ambitious sewerage scheme is being implemented in the metropolitan area of Bilbao City (North Spain) in order to recover the environment of the highly polluted Bilbao estuary. To check the efficiency of this scheme, the performance of the sewer system and the water quality in the Bilbao estuary and beaches have been simulated during a future hypothetical bathing season. The system has been tested under dry and wet weather conditions, using selected historical rainfall records, in order to evaluate the effect of the faecal pollution arising from Combined Sewer Overflows on the water quality of the beaches located in the outer part of the estuary. The results obtained indicate that under dry weather and optimal operational conditions of the sewer and treatment works, the mandatory European standards on bathing waters will be met in all beaches. The discharges caused by storm overflows will increase the faecal contamination in the beaches compromising the compliance with the guideline standards. Therefore, a disinfection of the treatment works effluent, the main faecal discharge to the estuary, may be needed.

Introduction

The Metropolitan area of Bilbao City and its surroundings are located along the axis of the Bilbao estuary (Fig. 1). This area, with a total population of approximately 1 million and more than 2700 industrial facilities, represents the largest concentration of inhabitants and industry on the Atlantic coast of Spain. The whole pollution generated by this human activity has been discharged into the estuary leading to an intense degradation of this ecosystem. Severe eutrophication symptoms (i.e. oxygen depletion and loss of animal life) as well as aesthetic and health-related problems have been widespread in the estuary since the last century, especially in the inner reach (Sáiz-Salinas & González-Oreja, 2000).

The outer part of the estuary, considerably less polluted, exhibits an important recreational value due to the existence of three beaches, located on the right bank, which are widely used during the bathing season and a new marina facility. However, the faecal pollution coming from the mouth of the estuary impacts on the water quality of these beaches causing failure to comply with the European Union (EU) Directive on bathing waters quality (EEC, 1976).

To improve the environmental situation in the estuary, a Sewerage Scheme involving the collection, transport and treatment of the whole sewage produced in the watershed is being implemented. This work commenced in 1984 and is being undertaken by the Consorcio de Aguas Bilbao Bizkaia (CABB). This is a corporate association of local municipalities that manages both the water supply and the sewerage and sewage treatment for the Metropolitan area of Bilbao. The scheme is currently developing rapidly, driven by the need to satisfy the EU Directive concerning the collection and treatment of urban wastewater (EEC, 1991).

The sewer system is combined and is composed of 170 km of interceptors and a central wastewater treatment plant (Galindo WWTP). After implementation of all works planned, Galindo WWTP will be able to biologically treat the wastewater produced in the whole catchment under dry weather conditions. However, under intensive rainy periods and, due to the combined nature of the sewer system, combined sewer overflows (CSOs) will spill. This is likely to affect the water quality in the estuary and at the beaches compromising the compliance with the European Directive on bathing waters.

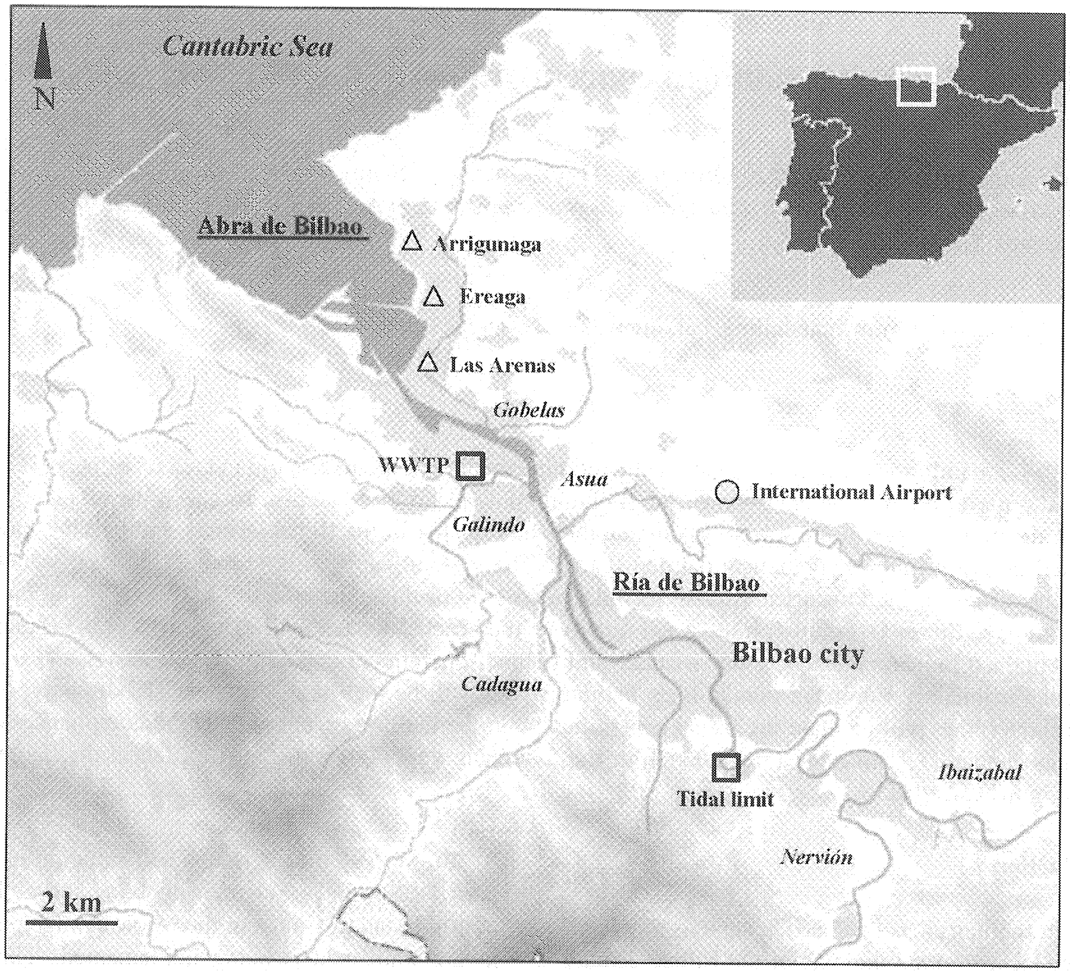

Figure 1. The study area showing bathing water compliance locations (△), the main river network and the location of the rainfall gauge at Sondika International Airport (○).

Because of the complexity of the processes involved, modelling represents a valuable tool to tackle this kind of problem. Recently, models have been extensively used in research as well as in the design and assessment of water quality management measures throughout the world (Rauch et al., 1998).

The CABB has started the studies to deal with the problems of wet weather discharges by means of mathematical modelling of the sewer system and receiving waters system using an integrated approach. In this report, we show an example of how modelling can be a useful tool to test the efficiency of the sewerage scheme in relation to current environmental legislation.

Study area

The Bilbao estuary is an urban mesotidal system with a semidiurnal tidal regime (Fig. 1). The system receives a moderate freshwater inflow from two main tributaries (Nervión and Ibaizabal) that converge 8 km upstream of the tidal limit; and from smaller tributaries along its way to the sea (namely: Cadagua, Asua, Galindo and Gobelas). The average flow at the tidal limit is about 25 $m^3 s^{-1}$.

In the estuary, two areas can be distinguished: a narrow, shallow and highly stratified channel (about 15 km long) that crosses the metropolitan area of Bilbao city (Ría de Bilbao) and a wide coastal embayment, with an area of about 30 km^2 called 'Abra de Bilbao'.

Situated on the left bank of the bay is the harbour and associated industry. By contrast, the right bank provides recreational amenity at their main beaches: Las Arenas, Ereaga and Arrigunaga (Fig. 1), which are identified under the bathing waters Directive for compliance monitoring.

The natural state of the Bilbao estuary has been modified by dyking large intertidal areas and by extensive dredging activities to maintain the navigation on the channel. In addition, the system has been subjecting to a huge loading of domestic and industrial wastes for over 100 years, leading to a severe degradation of this environment.

Material and methods

Modelling tools

A dynamic model of the sewer system has been developed using the 'Hydroworks' modelling system (Wallingford software). The model includes the network of main collectors constituting approximately 2200 nodes, 140 CSOs and serving 928 000 inhabitants. The total catchment area is 3700 ha, distributed into approximately 170 sub-catchments. This model is able to predict the water flow entering the sewer from storm water and to simulate the transport of sewage and associated pollutants through the sewer network.

The Ría de Bilbao is modelled with a one-dimensional, two-layered model (MIKE12 version 3.2, Danish Hydraulic Institute) in order to describe the stratified flow dominant in this part of the estuary. The upstream boundary is located at the tidal limit whereas the downstream boundary is located at the mouth of the estuary, in the Abra. The river network consists of one main river (principal channel), two tributaries (Cadagua and Galindo) and four closed basins. The rest of the tributaries are defined as lateral inflows to the main channel.

The model is composed of three modules: hydrodynamic, advection/dispersion and water quality. The hydrodynamic module simulates unsteady idealized two-layer stratified flows in a network of open channels. The results from this module consist of time series of water levels, interface levels, discharges, salinity and temperature for both layers. The advection/dispersion module describe the movement of pollutants in the water column and is based on the one-dimensional equation of conservation of mass. The water quality module, which is coupled to the advection/dispersion module, deals with the transforming processes of compounds in the water. The state of the system is described by different state variables (i.e. dissolved oxygen, ammonium, nitrate, phosphate, biochemical oxygen demand and coliform bacteria). This module solves the system of differential equations describing the physical, chemical and biological interactions between the state variables, calculating their concentration at each time step.The advection/dispersion and water quality modules run simultaneously and use the results generated by the hydrodynamic model as input data.

The Abra de Bilbao is modelled through a two-dimensional, one-layered model (MIKE21 version 2.7, Danish Hydraulic Institute). The upstream boundary is closed, with an artificial bathymetry which compensates for the volumes that enter/leave the estuary. The downstream boundaries at the open sea are tidal level boundaries.

The model includes hydrodynamic and particle movement modules in order to simulate the dispersion and decay of faecal coliforms in the sea.

Simulation strategy

The performance of the sewer system and the faecal coliform (FC) concentrations in the estuary and at the beaches have been simulated during a period of 4 months (covering a hypothetical June–September bathing season) under three different meteorological conditions. The first scenario consisted of a dry weather situation (i.e. no rain during the period of simulation). In the second scenario, historical rainfall records were used to simulate the effect of the pollution arising from storm overflows on the water quality of the estuary and at the beaches.

The rainfall events used consisted of monthly averaged rainfalls recorded in the period from 1967 to 1987 at Sondika International Airport, located a few kilometres from Bilbao city (Fig. 1). Finally, in the third scenario, the effect of a disinfection of the Galindo WWTP effluent under wet weather conditions has also been evaluated.

A constant river flow representative of summer conditions (5 m^3 s^{-1} for the Nervión-Ibaizabal system) has been used. The effect of the wind on the FC dispersion in the sea has not been considered. A constant FC decay rate (T_{90}) value of 16 h has been used for the marine area during the whole period of simulations.

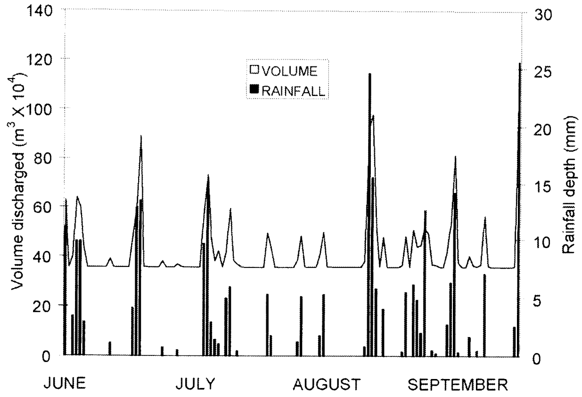

Figure 2. Daily volume of sewage discharged to the estuary as calculated by the sewer model. The baseline represents the dry weather flow. The solid bars represent the rainfall records used as input data.

The sewer model, the river/estuarine model and the marine model have been used sequentially, that is, the results from one model have been used as input data for the next model.

Results

After the full implementation of the sewerage scheme, under dry weather conditions, all the sewage produced in the area surrounding the estuary will receive a biological treatment before discharge through Galindo WWTP. The average dry weather flow will be approximately 350 000 m^3/d.

The sources of faecal pollution in the estuary include the outlet of the treatment works, which will represent about 98% of the whole FC load, and the pollution entering from the tributary rivers (2%). In addition to these continuous discharges, under intensive rainfall conditions intermittent discharges from CSOs will occur at many places along the estuary.

According to the sewer model predictions, the runoff generated by the rainfall records used produced overflow peaks in the sewer that represented about 18% of the dry weather flow (Fig. 2). These overflows were mainly discharged through the outlet of Galindo WWTP and produced an increase of about 26% of the dry weather FC load to the estuary.

The spill frequency through the CSO structures ranged between 8 and 51 spills per bathing season.

The wet weather discharges produced short-term (up to 20 h) marked increases in the FC concentrations in the estuary (Fig. 3). The magnitude of the observed FC concentration peaks was dependent on the amplitude and phase of the tidal cycle, and locally raised the FC concentrations up to 4–5 times the dry weather concentrations.

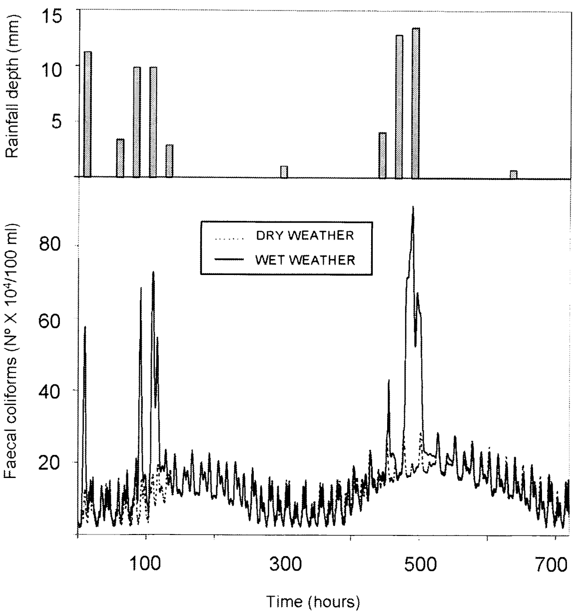

Figure 3. Simulated FC concentrations in the Bilbao estuary (11 km downstream of the tidal limit) during the month of June. The upper part of the Figure represents the rainfall used as input data for the sewer model under wet weather conditions.

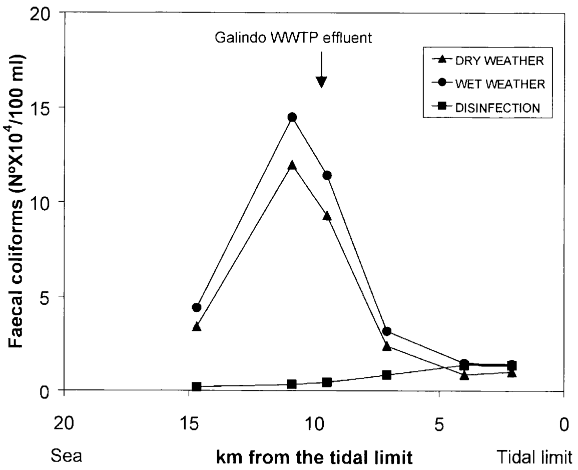

Figure 4. Averaged FC concentrations in the Ría de Bilbao calculated by MIKE12 under three different scenarios. The values represent the arithmetic means for the whole period of simulation.

Nevertheless, considering the whole period of simulation, the storm overflows produced, on average, an increase of the FC concentrations in the Ría de Bilbao of about 20% with respect to the dry weather situation (Fig. 4).

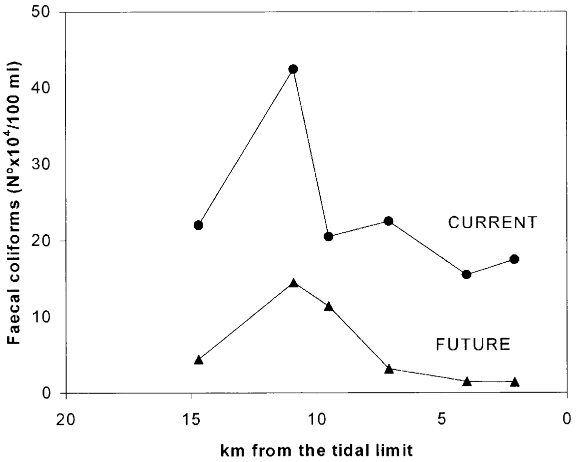

Figure 5. Current (real) *versus* future (simulated) FC concentrations in the Ría de Bilbao. The real values are averaged concentrations for the whole bathing season measured during the monitoring campaign carried out by CABB during 1999.

Table 1. Percentage of time exceeding the FC guideline value (100 FC/100 ml). Values estimated with MIKE 21 for the whole bathing season

Condition	Beach		
	Las Arenas (%)	Ereaga (%)	Arrigunaga (%)
Dry weather	76	1.75	2
Wet weather	78.5	3.75	3.75
With disinfection	1.6	1.3	0.8

The longitudinal profile of the coliform concentrations in this part of the estuary (from the tidal limit to the 'Abra') presents a maximum after the discharge of the Galindo WWTP effluent (km 10). The disinfection of this effluent would cause a dramatic reduction in the FC concentrations in the estuary, since this is the main source of faecal coliforms (Fig. 4).

Comparing to the current situation (CABB, 2000), the bacteriological pollution in the estuary (measured as FC counts) will be, on average, reduced by 75% after the completion of the sewerage scheme (Fig. 5). This is likely to produce a general improvement of the water quality of the Abra beaches.

The results obtained for FC show that, in the conditions simulated in this study, all the beaches would comply with the EU Directive (76/160/EEC) mandatory standard (95% of the samples can not exceed 2000 FC/100 ml). However, the guideline value (100 FC/100 ml, <80% of the samples) would occasionally be exceeded during the bathing season (Table 1).

The storm overflows produced, on average, an increase of the FC concentrations of about 25% with respect to the dry weather conditions. However, some CSO produced elevated concentration peaks of short duration up to ten folds the guideline threshold (i.e. 1000 FC/100 ml).

Taking into account the whole period, Las Arenas beach (the closest to the mouth of the estuary) would fail to comply with the guideline value under both dry and wet weather conditions. However, the disinfection of Galindo WWTP effluent would reduce the FC concentrations significantly reducing the time during which guideline values are exceeded (Table 1). The beaches located in the outer 'Abra' (Ereaga and Arrigunaga) would be less impacted by the faecal contamination coming from the 'Ría'. Although some local storm overflows directly affect these beaches, increasing the coliform concentration above the guideline value, on average, the exceedance period would only be about 3% of the bathing season (Table 1).

Comparing the current sanitary conditions of the Abra beaches (CABB, 2000) with the situation forecasted in this study, there would be a clear improvement after the completion of the sewerage scheme (Fig. 6).

Currently (1999), the faecal coliform concentration at the beaches exceeds the mandatory value during 50% of the bathing season and is above the guideline value for 95% of the bathing season (Fig. 6).

According to the results shown here, in the future, the three beaches will be compliant with the mandatory value and only Las Arenas beach would fail to comply with the guideline value (Fig. 6). Therefore, a disinfection of Galindo treatment plant effluent may be needed.

It should be stated that the riverine and marine modelling used in this study need to be improved to some extent (i.e. some calibration parameters must be tuned up). Therefore, the results shown here should be considered as a first attemp to predict the future sanitary conditions at the beaches after the completion of the sewerage scheme. Refinement of the modelling will be the subject of future research at CABB.

Discussion

The effect of future faecal coliform loads to the Bilbao estuary on the water quality at the beaches located

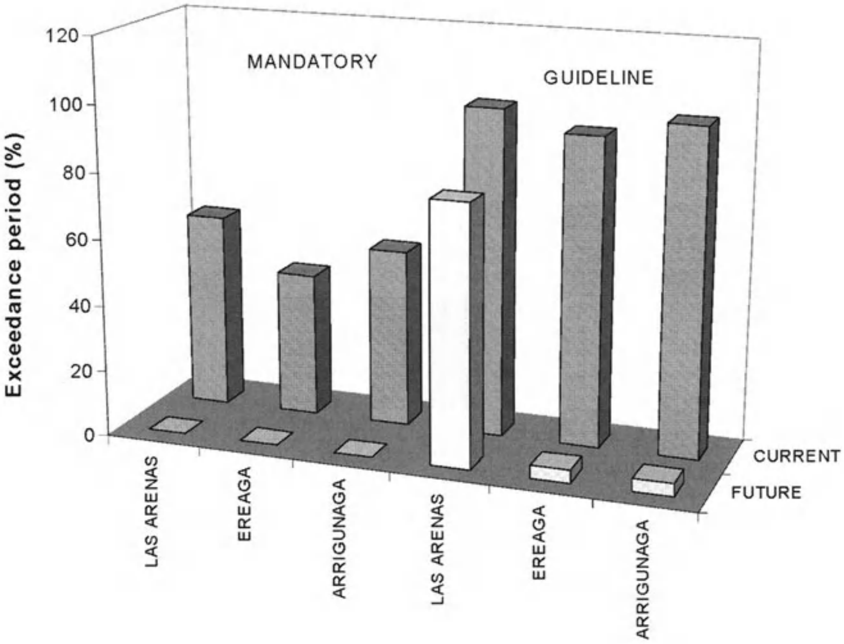

Figure 6. Current (real) *versus* future (simulated) mandatory and guideline values exceedance periods. The exceedance period is expressed as percentage of time of the bathing season. Real data come from the bathing waters monitoring campaign carried out by CABB during 1999.

at the estuary mouth has been evaluated (in terms of compliance with the EU Directive, 76/160/EEC) by means of mathematical modelling, combining sewer and estuarine/marine models.

The simulations carried out in this study indicate that the sewerage scheme will be efficient in reducing sewage pollution at the beaches to a level enough to allow compliance with the EU Directive on bathing waters.

In order to reduce complexity in this study, some simplifications that might lead to an underestimation of the real FC concentrations have been made, due to model limitations (i.e. consideration of a constant river flow under wet weather conditions, neglecting of the FC dispersion caused by the wind). Nevertheless, using a quite long FC survival time (T_{90}) compensates the effect of these assumptions. The T_{90} considered in this study (16 h) may be somehow high if compared with the very short T_{90} (0.5–2 h) values reported for the Cantabrian sea in previous studies (Canteras et al., 1995). However, T_{90} values as high as 7 days have been reported for turbid and organic-enriched estuaries (Salomon & Pommepuy, 1990).

It is well established that sunlight radiation is the main factor controlling the survival of faecal coliforms in seawaters (Solic & Krstulovic, 1992). We consider 16 h as a reasonable T_{90} for our studies due to (i) the high turbidity of the Bilbao estuary which will reduce sunlight penetration in the water column and (ii) the elevated number of cloudy days during the bathing season which would favour bacterial survival.

A considerable amount of faecal coliforms will be discharged daily to the estuary, mainly through the effluent of the Galindo WWTP. However, the impact of these discharges on the bacteriological quality of the beaches would be, on average, minimal as predicted by the models. This fact may be explained by the large distance from the point of discharge (Galindo river) to the beaches (6–10 km) and the high dilution provided by mixing with seawater.

It should be pointed out that even after the interception and treatment of all sewage discharging to the estuary, non-outfall and, in some cases, non-sewage sources of FC (diffuse pollution) may jeopardise the compliance with the bathing waters Directive.

The importance of these sources of faecal pollution, that have not been taken into account in this study, has been previously described (Wyer et al., 1997, 1998).

Wet weather impacts arising from urban catchments are widely recognised as a major cause of unsatisfactory receiving water quality (FWR, 1998).

This problem is especially important in coastal zones where the number of rainy days during the bathing season is high, like the area of study of this work. A surprising result of this study is the high occurrence of spills from CSOs (252 spills/bathing season) during wet weather, indicating a limited capacity of the sewer system. In spite of the high frequency, the impact of the storm overflows would be mainly aesthetic, since the increase in the faecal coliform concentrations at the beaches would not prevent the compliance with the EU Directive regarding the FC mandatory value. Therefore, a mechanical pretreatment of the CSOs to remove gross solids before discharging might be satisfactory in order to strictly comply with the mandatory value of the bathing waters Directive.

Nevertheless, if the guideline instead of the mandatory value were to be met, a disinfection of Galindo WWTP effluent would be necessary. The removal of this faecal pollution source, at least during the bathing season, would add an important safety margin in the compliance with the Directive.

In addition, there is a growing need to modify the current Directive as it shows serious limitations, being the most important one that it largely fails to reach the principal objective of reducing public health risk. In fact, on several occasions, the monitoring is made once the public has been exposed to risk. On the other hand, sometimes the risk is not real due to the transitory nature of many pollution events. To overcome these and other shortcomings, a classification scheme, based on health risk, has been recently proposed in which a beach would be assigned to a class (i.e. very poor, poor, fair, good or excellent) (WHO, 1998).

Therefore, the emphasis of any new Directive will be on managing bathing waters to protect public health rather than monitoring water quality to detect compliance. In that sense, the objective of any sewerage scheme regarding the microbiological quality of receiving waters would be, not only to guarantee compliance with the standards but to reach the best allowable quality level in a cost/benefit manner.

The present study shows the capability of modelling to address environmental problems that are governed by complex processes, such as the FC dispersion in near-shore waters. The use of models is becoming a widespread practise in the planning phase of many sewerage projects around the world (FWR, 1998). Furthermore, models could also be used as a powerful predictive tool in managing bathing waters. Modelling allows the simulation of as many scenarios as required taking into account both constant and intermittent inputs to bathing waters and their impact under different tidal and hydrometeorological conditions.

This predictive modelling approach would not replace the necessary *in situ* monitoring, but would help to identify pollution levels during special event conditions and to prevent bathers from being exposed to health risk before sampling results are available.

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Simulating vertical water mixing in homogeneous estuaries: the SADO Estuary case

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Key words: estuaries, numerical models, vertical transport, generic mesh

Abstract

In this article, a three-dimensional primitive equation model is applied to Sado Estuary, which is a meso-tidal homogeneous estuary. Despite the homogeneity of its density field, the flow presents important vertical structures. This article shows that the common practice of using two-dimensional depth-integrated models in homogeneous estuaries can filter important vertical hydrodynamic features, such as secondary and circulating flows. The physical processes associated with these flows are analysed. The strong curvature of the channels and the strong bathymetry gradients were identified as the principal morphologic features of the estuary responsible for the generation of the flows. The model possesses a generic vertical mesh geometry approach, which is used to test the sensibility of the model to the vertical discretization when simulating these flows.

Introduction

Mixing between bottom and surface estuarine waters is strongly dependent on the processes that occur in the vertical direction. This applies both for stratified and homogeneous estuaries. The use of three-dimensional (3D), primitive equations hydrodynamic models is usually restricted to estuaries with significant density stratification. In density homogeneous estuaries the common practice is to use simpler two-dimensional (2D), depth-integrated models. In stratified estuaries the need for a 3D model is easily understood since density gradients induce depth-varying horizontal forcing, producing a 3D flow pattern (Haney, 1991). Besides this direct effect, density gradients also influence the vertical transport of momentum, controlling the vertical eddy viscosity and thus the flow structure (Mellor & Blumberg, 1985). In density homogeneous estuaries, the density gradients do not play any role in the forcing but there are morphologic features of the estuary that can induce important 3D flows. The use of 2D models in estuaries with these characteristics will filter the vertical flows producing important errors in the vertical transport (Langerak, 1987). In this work, a 3D hydrodynamic and transport model is applied to Sado Estuary to identify the morphologic features responsible by the 3D flows and to study their impact on the transport of tracers. The conclusions obtained for Sado Estuary can be generalized to other homogeneous estuaries exhibiting the same characteristics.

Sado Estuary description

Sado Estuary is located 40 km South of Lisbon, Portugal. It is about 20 km long and 4 km wide as shown in Figure 1. The maximum depth is larger than 50 m and average depth is about 8 m. Inter-tidal sandbanks individualize a North and a South channel near Setúbal city. The channels display strong curvature near the mouth of the estuary. On the upper reaches, inter-tidal mudflats and salt marshes occupy about 1/3 of the estuary.

The tide is semidiurnal. The amplitude is about 1.6 m in spring tides and 0.6 in neap tides. The most important tidal components are M2 and S2. At the mouth, their amplitudes are 0.98 m and 0.35 m, respectively, both being amplified inside the estuary. The river flow displays a strong seasonal variability. Monthly average values can be as low as 1 $m^3\ s^{-1}$ in the summer months and values of 1800 $m^3\ s^{-1}$ can occur in the winter. The

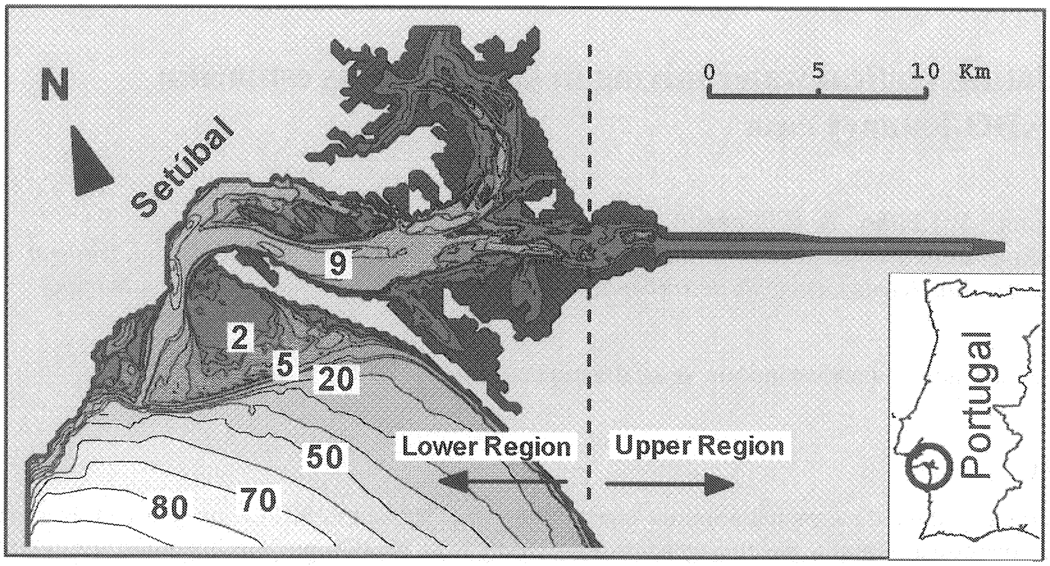

Figure 1. Sado Estuary bathymetry (meters).

annual average is about 40 m^3 s^{-1} (Vale et al., 1993). Flow is mainly tidally driven and strong residual eddies are produced (Martins et al., 2001). The estuary is subdivided into upper and lower parts as suggested by Wollast (1979), based on the temperature and salinity distributions. The lower estuary behaves as a coastal lagoon with small fresh water influence, while the upper river channel behaves as a typical long and narrow estuary.

A navigation channel 14 m deep is dredged across the platform located off the estuary. This channel intensifies the ebb stream issuing from the estuary's mouth and modifies the flow pattern in the adjacent slope between the 5 m and the 20 m isobaths.

Model and simulation descriptions

The model used is a 3D, primitive equation, finite volume model. The vertical discretization is based on the hydrostatic hypothesis, using the Boussinesq approximation. The vertical mesh geometry is of the generic type, allowing the use of any user defined law as well as traditional mesh laws such as Sigma (Phillips, 1957) and Cartesian (Bryan, 1969). For a more detailed description of the model, see for example Martins et al. (1998).

One of the main objectives of this simulation is to characterize the structure of the vertical flows present in Sado Estuary and the importance of these flows to the vertical transport. The sensibility of the model to vertical mesh geometry and viscosity is also an important target. An eulerian transport model is used to transport a passive tracer with a defined initial distribution. The study is focused in processes occurring on the slope close to the estuary bar. During most of the year, that region is characterized by a strong pool of nutrients for a depth greater than 10 m (Brogueira et al., 1994). For that reason, the transport model is initialized with a passive tracer possessing a value of zero for depths above 10 m and a value of 100 for depths below this value. The model is forced by tidal elevation on the open boundary. The tidal harmonics of the Setúbal tide-gauge are used, corrected in phase for the boundary location. The wind wave impact inside the estuary is small, for this reason wind waves are not considered. Wind stress is also not considered. Due to the small river runoff, the model is run in barotropic mode, thus neglecting density forcing. The effect of thermal stratification on the vertical eddy viscosity is however included in one of the runs. The bathymetry used in the 3D-model cover the lower estuary region as mentioned in the previous section. The model is coupled to a 1D model for the upper region. This is important in Sado since the tide signal travels for more than 40 km inshore.

Results

The model was previously calibrated and validated for Sado Estuary using measurements from 6 tide gauges

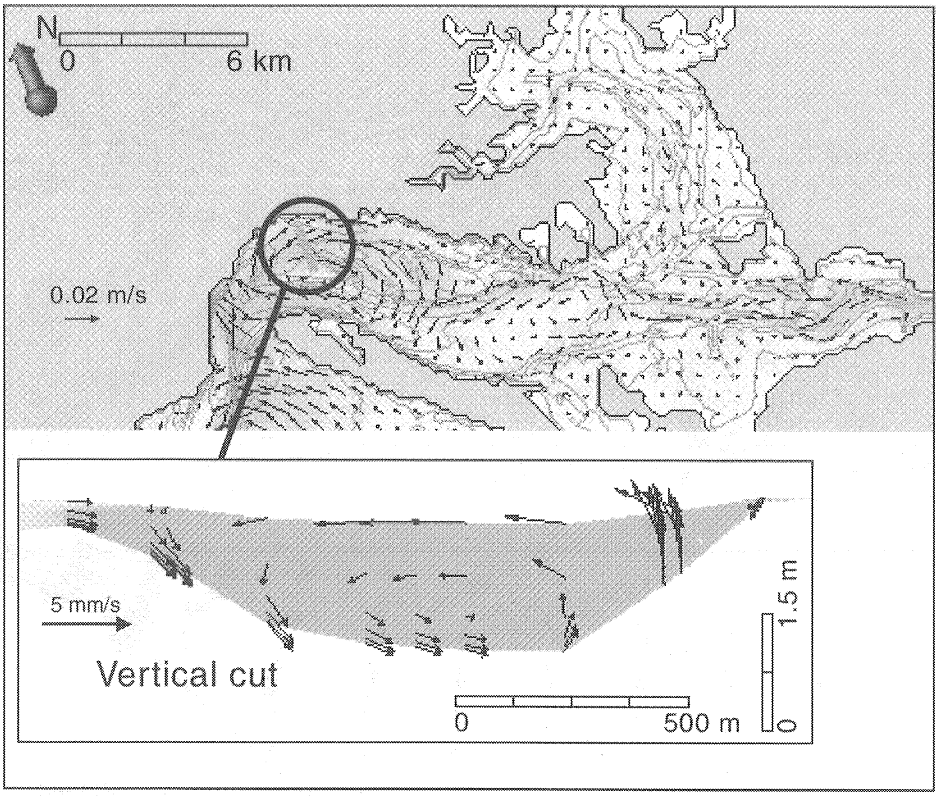

Figure 2. Eulerian averaged residual flow on the top layer for a neap-spring period (top); residual secondary flow on a vertical plane through the North Channel (bottom).

and 14 current meters in several points inside the estuary (Martins et. al., 2001). The model was run for a neap-spring period using the calibrated configuration with 6 sigma layers. Instantaneous and eulerian averaged residual velocities were computed. The passive tracer field evolution was also computed in this run.

Markedly 3D flow patterns can be identified both in the instantaneous and in the residual velocity fields. In Figure 2, a secondary flow can be identified in a vertical cut through the North Channel. In this case, the residual flow field is shown. This type of flow is associated with the curvature of the channel as explained in the next section.

A cut through the slope region offshore the estuary's mouth is shown in Figure 3. A recirculation eddy can be identified in this cut. This eddy lasts for almost 3 h of the ebb period, being destroyed during the flood. This type of flow is associated with the high bathymetry gradient of the region and with the ebb jet, enhanced by the navigation channel.

To investigate the impact of the vertical eddy viscosity closure scheme on this flow, several runs were made with different viscosity values and turbulent closures. The velocity field 4 h after high water for different viscosity values is shown in Figure 4. The top frame in this figure was obtained using the calibrated run and is here considered as the reference situation. The recirculation is more intense for lower vertical transport rates of momentum as explained in the next section. The model was also run with a turbulent closure of the mixing length type, taking into account the thermal stratification of the region. The results (not shown) did not differ significantly from the reference run. This shows that the typical stratification degree of this region is not strong enough to destroy the recirculation flow pattern.

The use of a generic mesh geometry model enables the use of different mesh geometries with the same code. The mesh geometry is closely related to the vertical transport of momentum via numerical diffusion (Mellor & Blumberg, 1985). In Figure 5 three runs using different mesh geometries are presented.

The vertical viscosity reference value was used and the time instant of the pictures is, as in Figure 4. The

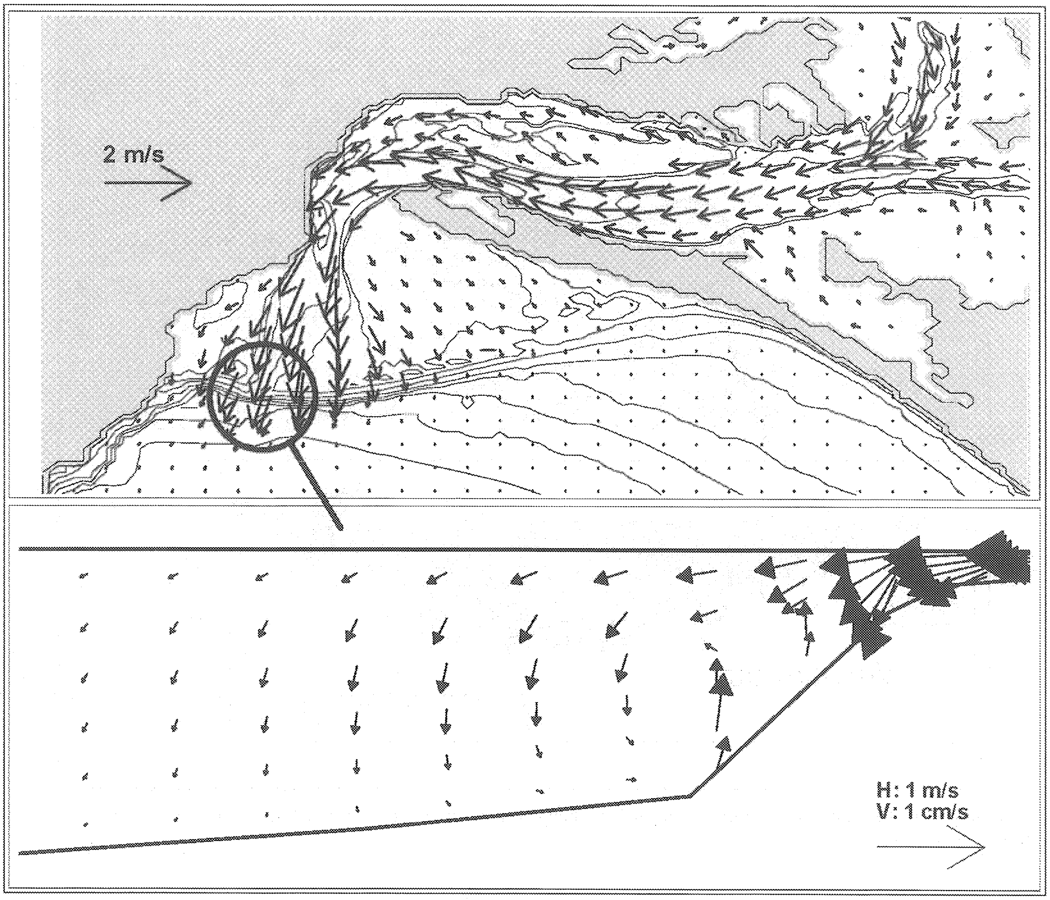

Figure 3. Instantaneous flow on the top layer 4 h after high waters (top); instantaneous recirculation eddy on a vertical plane normal to the slope (bottom).

lagrangian mesh type shown in the top frame was initiated as a sigma mesh and the layers were moved based on the vertical velocity field. Several methods were tested with this type of geometry to prevent excessive deformation of the mesh. Dumping the movement of the layers for thickness below a critical value gave the best results. The velocity fields obtained with this mesh are qualitatively similar to the reference sigma run (top frame in Figure 4). The sigma geometry is known to be well adapted to this type of simulation (barotropic with important depth variations). The similitude of the results thus shows the good behaviour of the lagrangian mesh for these conditions. This type of mesh is expected to behave better than the traditional sigma mesh in baroclinic stratified domains. In the center frame of Figure 5, the results for a Cartesian geometry with the same number of layers are shown. This configuration does not represent adequately the recirculation due to excessive vertical numerical diffusion. To overcome this problem a larger number of layers is needed as shown in the bottom frame of Figure 5.

In the slope region, the depth passes abruptly from about 7 m to more than 30 m. The recirculation eddy, therefore, penetrates the conceptual pool of nutrients, represented here as a pool of passive tracers initialised to a depth of 10 m. The transport of the tracer was started at high waters using the reference run conditions.

The tracer field, 6 h after high waters, is shown in Figure 6. The recirculation eddy produces a vertical transport of the tracer during the ebb period. At the end of the ebb the tracer already reaches the surface. This process influences the subsequent transport of the tracer over the platform during the flood. The tracer field for the top layer 6 days after the beginning of the run is shown in Figure 7.

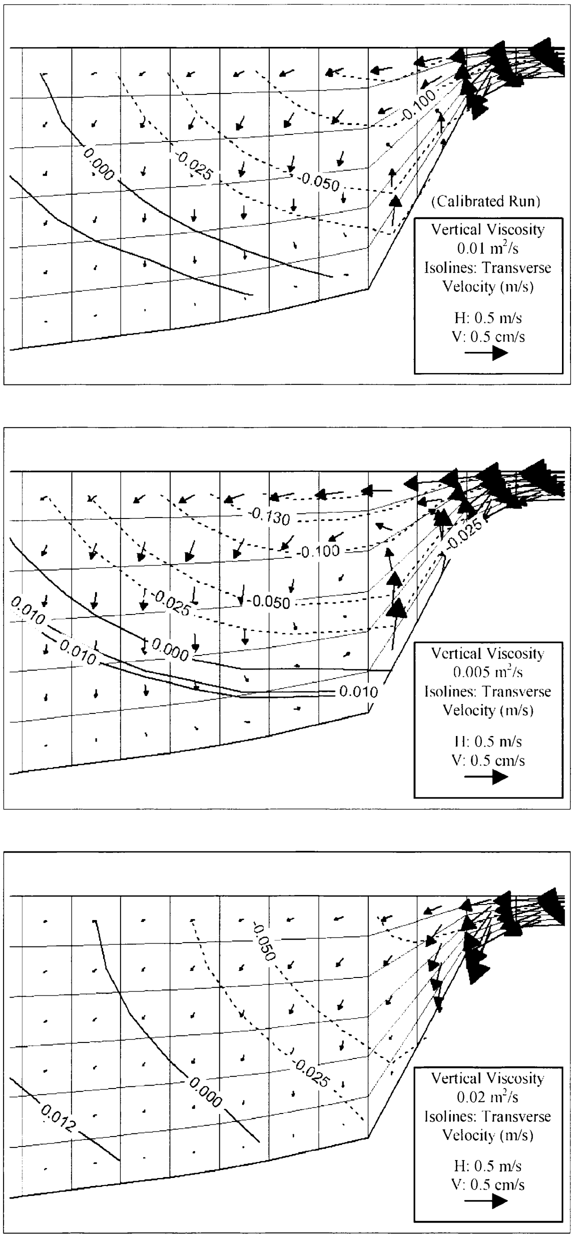

Figure 4. Recirculation sensitivity to eddy viscosity; reference run (top), half the reference viscosity (center), double the reference viscosity (bottom).

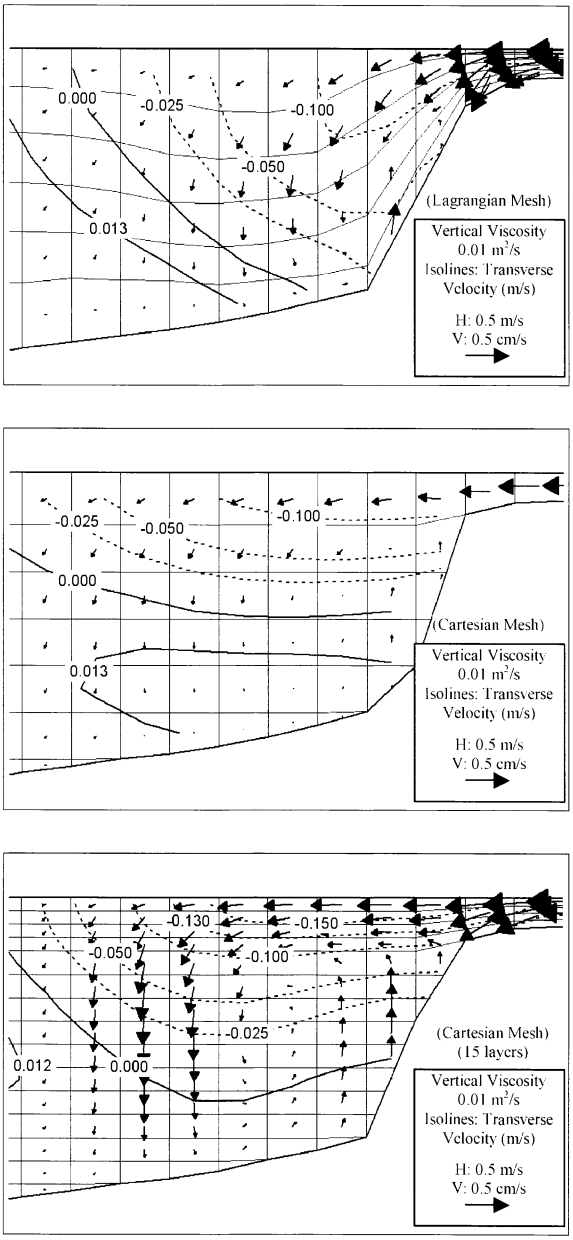

Figure 5. Recirculation sensitivity to mesh geometry; lagrangian mesh (top), cartesian mesh with the same number of layers (center), cartesian mesh with 15 layers (bottom).

Discussion

In this section, the physical mechanisms responsible for the 3D flows in this homogeneous estuary are analysed. The secondary flow presented in Figure 2 is produced by the horizontal pressure gradient due to the channel's curvature as will be explained with the aid of Figure 8. The water's free surface rises on the outside of the curve to compensate the centripetal acceleration produced by the curve. Since the system is barotropic, the pressure gradient produced by this surface tilt is equal at any depth. The centripetal force on the other hand is proportional to v^2/r were v is the water velocity and r is the streamline curvature radius. Due to shear, the velocity is lower near the bottom. Since the

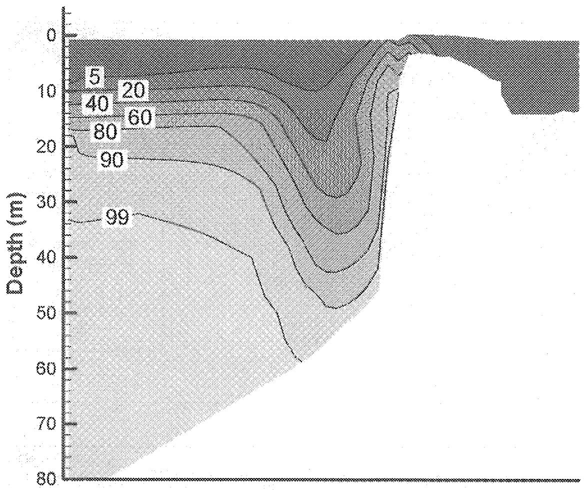

Figure 6. Vertical cut of the tracer field over the slope, 6 h after high waters.

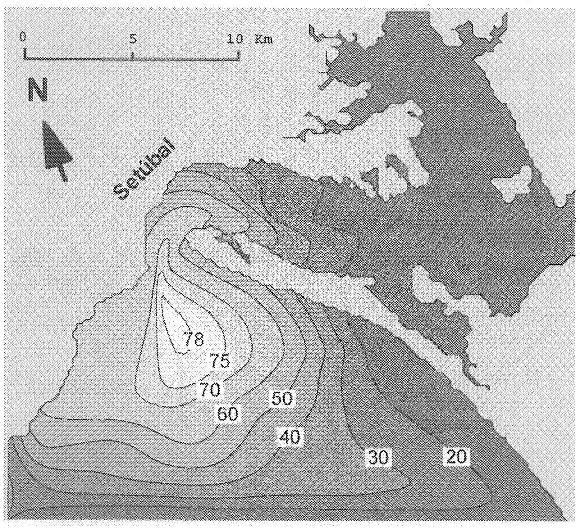

Figure 7. Horizontal tracer distribution on the top layer after 6 days.

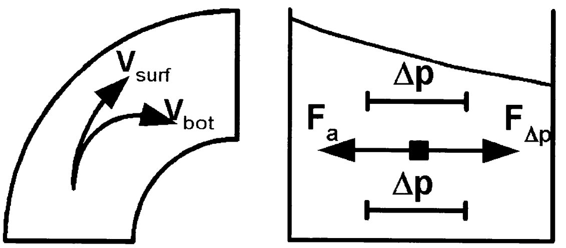

Figure 8. Physics of secondary flows: same pressure gradient for all depths (right); same centripetal acceleration implies higher curvature of bottom streamlines (left).

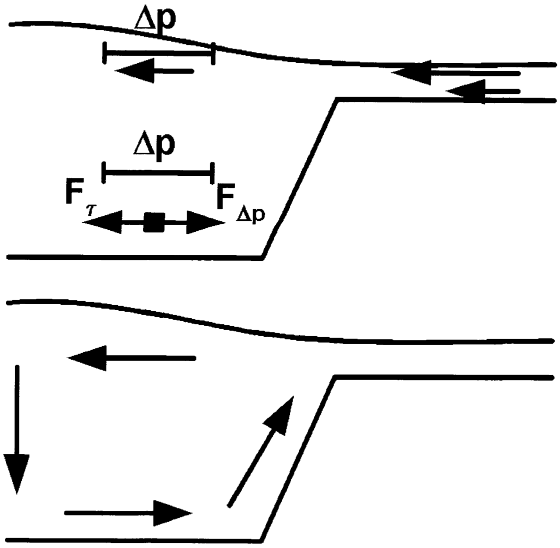

Figure 9. Physics of recirculation flows: same pressure gradient for all depths (top); vertical transport of momentum controls recirculation (bottom).

centripetal force must be the same to compensate the pressure gradient, the curvatures of the streamlines at the bottom are higher than at the surface. On a radial plan, this produces a flow towards the inside of the curve at the bottom and, by continuity a secondary flow is formed. This process is not dependent on the density gradient thus occurring in homogeneous estuaries. These flows can produce an important vertical transport and are completely lost when 2D Models are used.

The recirculation flow presented in Figures 3–5 is produced by the combined action of momentum and pressure as will be explained with the aid of Figure 9. During ebb, the depth increase of this step-like feature produce an increase of water level due to conversion of kinetic energy into pressure. Due to the barotropic nature of the system, this horizontal pressure gradient have the same magnitude at all depths. The bottom flow possesses a velocity lower than the surface flow being retarded in a more significant way by the pressure gradient. On the other hand, the vertical diffusion of momentum will produce on the bottom flow a force with the downstream direction. If this viscous force is not strong enough to compensate the pressure effect the bottom flow will reverse, producing by continuity a recirculation behind the step. The vertical transport of momentum controls the degree of recirculation, intensifying the eddy when the transport of momentum is low. This explains the flow sensibility to vertical viscosity as shown in Figure 4. The navigation channel enhances the ebb jet issuing from the platform, contributing to the growth of this recirculation. During flood the recirculation is destroyed by the incoming flow.

The above-mentioned phenomena are not particular features of this estuary; on the contrary they are likely to occur in a large number of homogeneous estuaries displaying strong curvature and bathymetry gradients. To accurately simulate water quality and primary production, the identification of these flows with a 3D hydrodynamic model is, therefore, an important step.

Conclusions

The 3D flows identified in this paper were shown to be important features of Sado Estuary. These flows are completely lost if a 2D depth integrated model is used. The physical principles behind them suggest these features may appear in other homogeneous estuaries when the curvature of the channels is high or when the batimetry gradient is important. In that cases, the vertical flows must be considered when the transport in the vertical direction is an important aspect. That is the case when ecological models are used. In the future, an ecological model will be applied to further investigate the impact of these structures on the transport of nutrients and on the production of the region.

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Mathematical modelling as a management tool for water quality control of the tropical Beberibe estuary, NE Brazil

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Key words: water quality, modelling, estuaries, QUAL2E, MUDLARK, Brazil

Abstract

Sewage disposal in natural waters is a common problem in most countries. Large inputs of organic matter and nutrients from raw sewage to a weak hydrodynamic environment may lead to deterioration of the water quality. Widely available riverine and estuarine models such as QUAL-2E and MUDLARK can be used to adequately model these situations. Beberibe is a low hydrodynamic estuary that runs through a densely populated region in the Recife Metropolitan Area (RMA), Northeast Brazil, and receives untreated domestic sewage from aproximately 200 000 inhabitants. The mouth of the estuary is shallow and tortuous, causing a large reduction in tidal propagation. The low river flow is strongly influenced by rain seasonality at the upstream boundary, with mean values varying from 4.9 m^3 s^{-1} in the wet winter to 1.4 m^3 s^{-1} during the dry summer. A major program to build sewage plants was planned to increase the water quality of the RMA rivers. This study focuses on water quality modelling of the Beberibe estuary basin, formed by the Beberibe River and two small tributaries. Numerical simulations of temperature, dissolved oxygen, biochemical oxygen demand, nitrate, ammonia, phosphate and faecal coliforms were carried out, targeting the expected population growth in the following 20 years. The QUAL-2E and the MUDLARK models were coupled at the tidal intrusion limit, with the estuarine sector modelled by the MUDLARK while QUAL-2E was used in the upper river. A longitudinal dispersion coefficient related to tidal excursion was introduced into the MUDLARK algorithm to better determine the tidal effect on the distribution of water quality variables. Both models were calibrated successfully and verified with a 4 year water quality data series from the Pernambuco State Environmental Agency (Companhia Pernambucana do Meio Ambiente – CPRH). Results showed that the river flow is a major factor controlling the water quality. Even the most efficient treatment applied was not able to bring water quality up to all desirable levels during dry summer months, mainly considering dissolved oxygen and biochemical oxygen demand. Results also confirm that the spring-neap cycle does not significantly affect water quality, probably due to the strong tidal attenuation at the estuary mouth.

Introduction

A great proportion of the world population lives close to estuaries, promoting large environmental changes, from shape modification caused by landfills and dredging to water quality deterioration. Apart from cities where the alternative of oceanic outfall has been used, estuaries are the most common place to dispose of domestic and industrial sewage from adjacent urban areas. Different degrees of treatment of the wastewater can be applied depending on the desirable final condition of the estuarine water quality. It follows that a cost-effective analysis and the applied treatment have to take into account the load characteristics and the natural dilution capacity of the estuary.

Numerical modelling is a useful tool to help decision making about sewage treatment configuration in order to achieve the targeted water quality. Different water quality models have been used successfully for a long time in several different estuarine systems (Bowie et al., 1985; Rauch et al., 1998). A broad variety of water quality models are available,

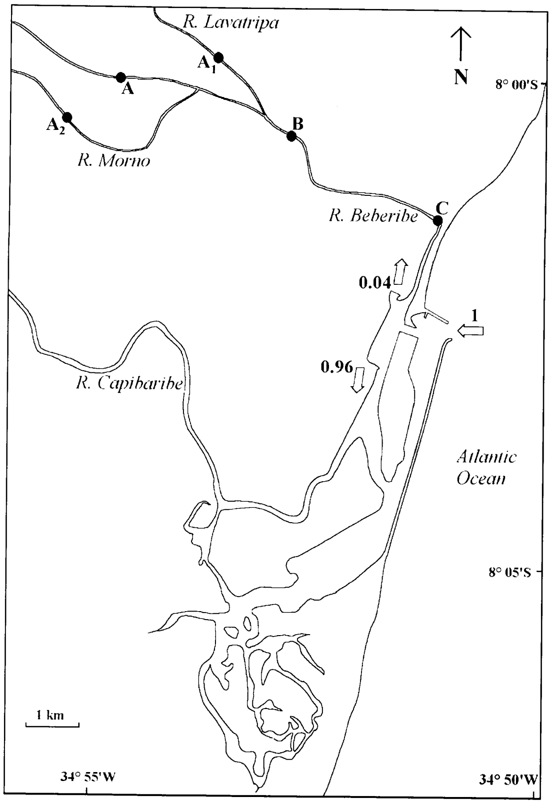

Figure 1. Capibaribe-Beberibe estuarine system, showing the tidal prism diversion southward to the Capibaribe basin. Unit was considered as total tidal prism incoming from the ocean. Modelling boundaries are named as A (Beberibe headwater), A_1 (Lavatripa headwater), A_2 (Morno headwater), B (tidal limit) and C (most downstream boundary).

from simple one-dimensional solutions with only the more important water quality parameters to complex three-dimensional schemes (Van Leussen & Dronkers, 1990). The choice of the numerical method depends basically on the characteristics of the system and on the computing time cost.

Here we decided to couple two simple numerical models to simulate different hydrodynamic situations in a small and highly impacted estuary, the Beberibe estuary. This work was motivated by a governmental plan to build four small sewage treatment plants at the Beberibe water basin, as part of a major sanitation program to benefit all 2.5 million people in the Recife urban area of Northeast Brazil. The aim of this paper is to assess relevant improvements on water quality of the Beberibe estuary after the implementation of the sewage treatment.

Description of the area

Beberibe is a 15-km long river that drains an area of approximately 150 km^2 through a highly populated region in the Recife Metropolitan Area (RMA), Northeast Brazil, located between latitudes 7° 59′ and 8° 02′ S and longitudes 34° 52′ and 35° 00′ W (Fig. 1). River flow is very low, varying from 1.4 to 4.9 m^3 s^{-1} in the dry summer and the wet winter, respectively. These values were averaged from a 4-year (from 1994 to 1997) daily measurement dataset taken in the upper third of the river (point A in Fig. 1). This low river discharge is due to the small catchment area. The regional rainfall regime is also greatly influenced by the El Niño Southern Oscillation (ENSO). For example, during the last 1998–1999 El Niño event, annual mean rainfall dropped from the 30-year mean of 1074 to 544 mm yr^{-1} in 1998 and 635 mm yr^{-1} in 1999. This reduction in precipitation caused severe droughts in the region and drastically diminished river flow. The semi-diurnal tidal range varies from 1.0 m during neap to 1.6 m during spring tide at the most downstream point considered in the model (Point C – Fig. 1). Estuarine tidal limit is located 5.5 km upstream from the river mouth in a step bathymetric feature (Point B), and was considered fixed in the modelling process to both fortnightly tidal cycle and seasonal river flow variation. The water volume that enters through the narrow oceanic communication is mostly diverted to the deeper and larger adjacent Capibaribe estuary, dredged for port activities and localised in a lower altitude area compared to the Beberibe basin (Fig. 1). Tidal prism is also strongly attenuated in the lower Beberibe system because of its shallow and constricted shape causing an even larger reduction in tidal propagation into the estuary. Considering the overall tidal prism of the Capibaribe-Beberibe system, less then 4% of this volume reaches the Beberibe estuary due to diversion to the Capibaribe estuary and tidal attenuation caused by morphological constraints. The Beberibe estuary tidal prism was calculated according to model A presented by Luketina (1998) for both spring and neap tide, with resulting flux values averaged over one tidal cycle of 4.03 and 2.35 m^3 s^{-1}, respectively.

An existing primary treatment plant, the Peixinhos plant, discharges sewage into the estuary from 385 000 of the total 585 000 people living in the Beberibe basin. However, the other 200 000 people, mostly living in poor shantytowns without any urban planning, discharge untreated sewage directly into the system.

A governmental program proposes the construction of four small primary treatment plants including disinfection, with each outflow ranging from 0.059 to 0.093 $m^3\ s^{-1}$, to serve this unassisted population. This program aims mainly to develop current sanitation conditions by building sewerage facilities and so improve the water quality in the Beberibe estuary. Characteristics of these new plants, such as the total number, location and capacity, were decided by the governmental plan through a cost-effective analysis in respect of the location of unassisted population, wastewater pipeline length and availability of land to build the plants (CCD, 1998). Water quality was not taken into account in their analysis. Given the final design of the planned sewerage network, this present study aims to simulate through numerical models the final water quality for the Beberibe estuary and two small tributaries.

The models

Two 1D models were used according to the different hydrodynamic characteristics of the estuary. The QUAL-2E model (NCASI, 1985) was employed at the unidirectional fluvial section of the system (hereafter referred to as upper river) while the MUDLARK model (Binnie & Partners, 1993) was used in the estuarine sector, where tidal motion acts.

The transport of dissolved substances in rivers is governed by advection and turbulent diffusion. For the water quality study of the upper Beberibe river, which is dominated by longitudinal and temporal changes, further integration is possible along the width. The 1D advection-dispersion equation can be written as

$$\begin{aligned}\frac{\partial C}{\partial t} =& \frac{1}{A}\frac{\partial}{\partial x}\left(AD_{Lq}\frac{\partial C}{\partial x}\right) - \frac{1}{A}\frac{\partial}{\partial x}(AuC)\\ &+ \frac{\mathrm{d}C}{\mathrm{d}t} + \frac{s}{V},\end{aligned} \tag{1}$$

where A is the cross-sectional area (m^2), D_{Lq} is the longitudinal dispersion coefficient ($m^2\ s^{-1}$), C is the cross-section averaged concentrations of various constituents (kg m^{-3} for chemical state-variables or °C m^{-3} for temperature) and s/V is the ratio of the cross-section averaged concentrations change (kg $m^{-3}\ s^{-1}$ for chemical state-variables or °C $m^{-3}\ s^{-1}$ for temperature) due to conversion processes which are now a function of model parameters.

Equation 1 is integrated in QUAL-2E for subsequent river stretches within which complete mixing is assumed. This procedure leads to a sequence of interlinked reactors or boxes (and the application of the 'reactor principle'). Hence the mass balance can be expressed by

$$\frac{\partial(VC^t)}{\partial t} = Q_iC_i - QC^t + VG^t(C^t,\ P^t), \tag{2}$$

where C^t is the concentration (kg m^{-3}), C_i is the inflow concentration (kg m^{-3}), Q_i is the inflow ($m^3\ s^{-1}$), Q is the outflow ($m^3\ s^{-1}$), V is the box volume (m^3) and $G^t(C^t,\ P^t)$ is the conversion rate (kg s^{-1} for chemical state-variables or °C s^{-1} for temperature). For m number of boxes and n number of variable, $n \times m$ ordinary differential equations are solved. Such box models are frequently used (e.g. Beck & Finney, 1987; Rauch & Harremoes, 1996) due to the easier formulation of calibration and verification methods in comparison to partial differential equation model structures (Rauch et al., 1998).

Each box or segment is connected to the two other segments along two sides. The advection term is estimated by the Manning equation

$$Q = \frac{1}{n}AR^{2/3}I^{1/2}, \tag{3}$$

which describes the advective flux Q ($m^3\ s^{-1}$) as a function of the transverse section area A (m^2), the hydraulic radius R (m), and the dimensionless energy line steepness I. By assuming the Manning roughness coefficient, n, as 0.02 (Thimakorn, 1990), the advection term is driven purely by the river geometry. The values of longitudinal dispersion coefficients strongly depend on slope, morphology and roughness, and can be taken from empirical formulae (Fischer et al., 1979) or from *in situ* tracer measurements. In the narrow upper Beberibe river the dispersion coefficient D_{Lq} was expressed by the classical Elder (1959) formulation $D_{Lq} = K\mathrm{d}u_*$, where K is the dimensionless Elder constant 5.93, d is the channel depth (m), and u_* is the wall friction velocity (m s^{-1}) estimated from $u_* = \sqrt{gRI}$, where $\boldsymbol{g}$ is the gravitational acceleration (m s^{-2}).

QUAL-2E models temperature, dissolved oxygen (DO), 5-day biochemical oxygen demand (BOD), organic nitrogen, ammonia, nitrite, nitrate, organic phosphorus, phosphate, faecal coliform (FC) and chlorophyll-*a*. It contemplates the following kinetic processes: degradation of organic material, growth and respiration of algae, nitrification (considering nitrite as an intermediate product), hydrolysis of organic nitrogen and phosphorus, reaeration, sedimentation of

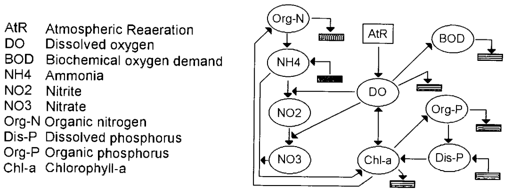

Figure 2. Schematic description of the kinetic processes on the water quality model QUAL-2E. Temperature and coliform are not represented here.

algae, benthic uptake of oxygen, and benthic flux of nitrogen and phosphorus (with their relations shown on Fig. 2).

All the processes in the QUAL-2E model consider the effect on oxygen, nitrogen and phosphorus cycles. Algae dynamics were not simulated in the present study because *in situ* estimations of DO production from photosynthesis were negligible when compared to DO reaeration inputs and to the magnitude of DO sinks at Beberibe system (Souza, 2000). Coliform bacteria modelling was carried out through the Schick's first order decay equation, and heat balance was taken into account through temperature state variable.

MUDLARK is a simple 1-D constant volume box model and, as QUAL-2E, is based on mass conservation equations. To keep the box volumes constant, the lengths of the boxes or segments stretch out and shrink into the estuary according to the segment depth changes ruled by the tidal range (O'Kane, 1980). The governing equations run for every time step during the whole tidal cycle, giving average results for the tidal situation. Lateral pollutant source and sink locations remain fixed and may, therefore, impact two or more segments during the tidal cycle.

Water quality parameters simulated in MUDLARK were temperature, salinity, DO, BOD, nitrate, ammonia, phosphate and FC. A longitudinal dispersion coefficient related to the tidal excursion was introduced into the MUDLARK algorithm to better determine the tidal effect on the water quality. It was adapted from a general expression proposed by Bowie et al. (1985) for an estuarine case as follows:

$$D_{Lm} = A_{\mathrm{d}} L^{4/3}, \tag{4}$$

where D_{Lm} is the longitudinal dispersion coefficient ($\mathrm{m}^2\ \mathrm{s}^{-1}$), A_{d} is the dissipation parameter ($\mathrm{m}^{2/3}\ \mathrm{s}^{-1}$) and L is the tidal excursion (m). The boundary between these two models was the tidal limit point. To couple the models, the most downstream results from QUAL-2E were considered as the headwater boundary input data to MUDLARK.

Simulations were carried out using different scenarios in order to understand how tidal and fluvial hydrodynamics affect the distribution of the water quality variables. As rainfall in the Northeastern Brazilian coast has a seasonal pattern, a 4-year dataset of daily river flow measurements was averaged within dry (September–February) and rainy (March–August) months. Beberibe river flow was measured at point A (Fig. 1), stated as headwater into the model. For the two small tributaries, Lavatripa and Morno, where outflows from two of the four planned sewage treatment plants are located, no previous river flow measurements were found. It was assumed that runoff in these two streams is the same as in the upper basin from the Beberibe river headwater, since there are strong similarities in the characteristics of the soil, superficial area, vegetation cover and land uses among all the three headwater drainage areas.

The governmental program plans to complete the sewage scheme implementation in 2010, therefore different population scenarios for years 2000, 2010 and 2020 were simulated to foresee the water quality benefit from the sewage treatment. The regional population growth rate of 0.5% yr^{-1} was used to calculate the expected population living in the basin for the next two decades. For each population scenario, there are four different hydrodynamic conditions according to the tide (spring and neap) and river flow (dry and rainy) boundary conditions. The planned sewage treatment including all four plants was then considered for the simulations for years 2010 and 2020. Simulations for 2000 were carried out just for the present situation with the discharge of raw sewage from the unassisted population and the current loads from the Peixinhos plant and the two industries in the Beberibe basin. This situation acted as a control to calibrate and verify the models with *in situ* measurements from a 4-year water quality dataset.

The overall stream length modelled was 14 km, with a spatial resolution of both models of 500 m, resulting in 28 well-mixed segments (Fig. 3). Bathymetric data were averaged for each segment. The tidal limit was placed 5.5 km downstream of the Beberibe headwater. Hence the QUAL-2E model was applied to the 21 segments of the upper river above the tidal limit, including the 4 km Morno and the 2 km Lavatripa streams, while simulations of the 3.5 km estuarine sector, i.e. 7 segments, were executed by the MUDLARK model.

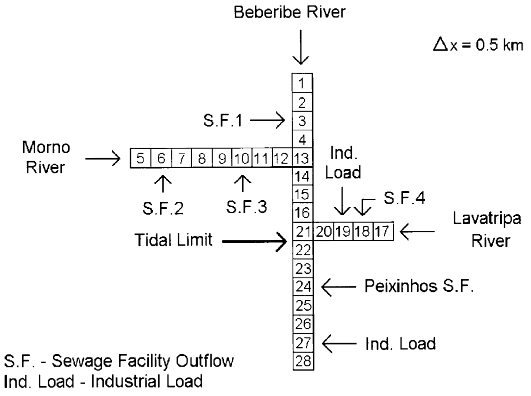

Figure 3. Conceptual diagram of the Beberibe basin modelling, showing the segmentation of the rivers considered on the models, the points of the sewage plants and industries outflow and the tidal limit.

Estimations of nutrients and BOD loads from untreated sewage were based on the unassisted population. The number of people living in the area draining into each segment considered in the models was calculated by overlaying density population and topography information. Inputs were obtained from the mean daily *per capita* load of 54 g of BOD, 6.7 g of ammonia, 1.7 g of phosphate and 2 × 10^9 MPN of FC, averaged from various estimates (Metcalf & Eddy, 1979; Hammer, 1986; Eckenfelder & Argaman, 1991; Jenkins & Hermanowicz, 1991; Klepper et al., 1995; Fernandes, 1997). To assess pollutant loads in each model segment, these estimates were multiplied by the population living in each segment drainage area. In spite of different decay ratio estimates to consider natural decay on the way to the stream (Klepper et al., 1995), the ratio of 40% was applied to the contaminants according to the local field experiments carried out by Souza (2000). The current outflows from the two industries in the Beberibe basin as well as the Peixinhos sewage treatment plant were also included. Air temperature, precipitation and relative humidity data were obtained from the closest National Meteorology Service station, 5 km south of the Beberibe basin.

A 4-year water quality monitoring plan was used to calibrate and verify both models. This monitoring plan has been executed on a monthly basis by the Pernambuco State Environmental Agency CPRH since 1994, sampling at five stations on the Beberibe river and one on the Morno stream. The variables measured and used here were temperature, conductivity, dissolved oxygen, BOD, nitrate, ammonia, phosphate

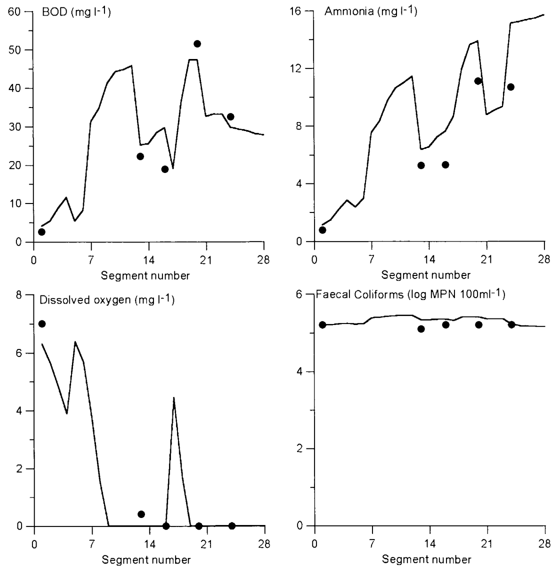

Figure 4. Calibration for BOD, ammonia, dissolved oxygen and faecal coliform, comparing the models results for the dry-neap non-treatment situation in all 28 segments modelled (full line) with averaged field observations from the 4 year monitoring plan in five stations (dots).

and FC. Data were averaged for wet and dry periods, after exclusion of outliers defined as $\bar{\theta} \pm 2\sigma_\theta$, where $\bar{\theta}$ is the mean value for the variable θ and σ_θ is the standard deviation. As the models simulated seasonally averaged situations, the removal of the outliers aims to have average conditions for dry summers and rainy winters avoiding values from somehow extreme events. At the only sampling station located on the tidal section, mean values were also calculated according to spring-neap cycle. Data from two stations were used as input for water quality variables to both the headwater and the most downstream estuarine boundaries in the Beberibe river, while calibration and verification were performed with the datasets from the other four stations. Analogous to the river flow evaluation already mentioned, it has been assumed that similarity between Beberibe river and the two tributary basins provides a reasonable approximation for the water quality at the headwater boundaries. Calibration and verification of both models were satisfactorily achieved for all water quality variables, allowing a maximum difference of 20% between model outputs and observational data (Fig. 4). The calibration of both models coefficients was accomplished by using the averaged summer dataset from four sampling stations for the dry season situation with the current sewage discharge situation, i.e. without the planned treatment

Table 1. Comparison of C_f/C_{sat} ratio for lower river flow during summer and higher river flow during winter, at the five treatment plants discharge points through the Beberibe basin rivers

Segment	Dry summer	Rainy winter
3	0.56	0.52
6	0.80	0.59
10	1.13	0.70
18	1.51	0.84
24	2.38	1.32

plants. After setting the coefficients for allowing a 20% error range for all parameters, winter averaged values were applied to verify the results by using rainy season conditions. This procedure was carried out continuously until the final results on the verification run were within the 20% error range limit.

The dilution factor

To indicate whether or not the flow is able to dilute the incoming sewage considering no kinetic processes, a simple mass balance equation can estimate the final concentration C_f of the BOD after the load dilution into the flow. Clark (1997) called it the dilution factor, defined as:

$$C_f = \frac{(Q_r C_r) + (Q_s C_s)}{Q_r + Q_s} \tag{5}$$

where Q_r and Q_s are the river flow and the sewage outflow (m^3 s^{-1}) respectively, and C_r and C_s are the river and effluent BOD concentrations (mg l^{-1}). To stress the difference between wet and dry seasons, mean river flow and mean river BOD concentration for rainy winter and dry summer were used. Comparison of the dilution factor C_f with the oxygen saturated water concentration C_{sat} indicates a quick but rough approximation of the necessary reoxygenation to oxidise the organic matter. It can be used to check critical sectors of the stream prior to more complex modelling. The C_f/C_{sat} ratio was calculated for the Beberibe system for all five segments that receive discharge from the current and planned sewage plants (Table 1). As most of the kinetic processes involving oxygen (e.g. nitrification, oxidation, respiration) are sink processes, C_f overestimates the BOD dilution capacity whenever photosynthetic activity is very low, as expected in extremely high turbidity waters like the Beberibe estuary.

Results

According to the results of the simulations using different hydrodynamic scenarios for the treated sewage condition, the difference between both river flow and tidal range regimes considered here promoted large modifications in the concentrations of the variables. The improvements in water quality were more significant during spring tides and higher river flow in rainy months. Water renewal may be greater during this more energetic period, leading to a higher dilution rate and lower retention time of the domestic sewage pollutants. It suggests that the water quality in the Beberibe system is extremely dependent on hydrodynamic conditions. However, river flow variability exerts a more effective control on water quality than the spring-neap tidal cycle. Both the pronounced tidal prism diversion to the adjacent Capibaribe basin and the attenuation in the first few kilometres of the Beberibe estuary work on reducing the influence of the tide on the whole water exchange balance.

A comparison between the lower and higher river flow for a non-treatment scenario and the current population highlights this strong hydrodynamic influence on the water quality (Fig. 5). Averaging the results for all 28 well-mixed segments of both models and considering spring tide conditions, dissolved oxygen doubled from 1.4 to 3.0 mg l^{-1} and BOD dropped from 25.0 to 12.2 mg l^{-1}, between lower and higher river flow. Averaged concentration of nutrients also decreased under higher flow conditions. Ammonia averaged concentrations fell from 9.70 to 3.77 mg l^{-1}, and phosphate from 2.13 to 0.78 mg l^{-1}, between low and high river flow. Reduction of the FC averaged values was less significant, from 3.07×10^5 to 2.62×10^5 MPN 100 ml^{-1}. Oxygen kinetic processes, which are intimately related to nutrients and DBO, are the probable causes to this less significant reduction of FC compared to the other pollutants during higher river flow periods, as FC are only temperature and time dependent. Variations of concentrations due to the spring-neap cycle reached less than 10% and were one order of magnitude lower compared to the seasonal river flow difference for all water quality variables.

The planned sewage treatment led to a significant reduction in pollutant concentrations (Fig. 5). Considering the 2020 population situation, averaged BOD concentration for all 28 segments dropped from 34.3 mg l^{-1} to 7.8 mg l^{-1} during low river flow and from 12.6 mg l^{-1} to 5.3 mg l^{-1} during rainy conditions, due

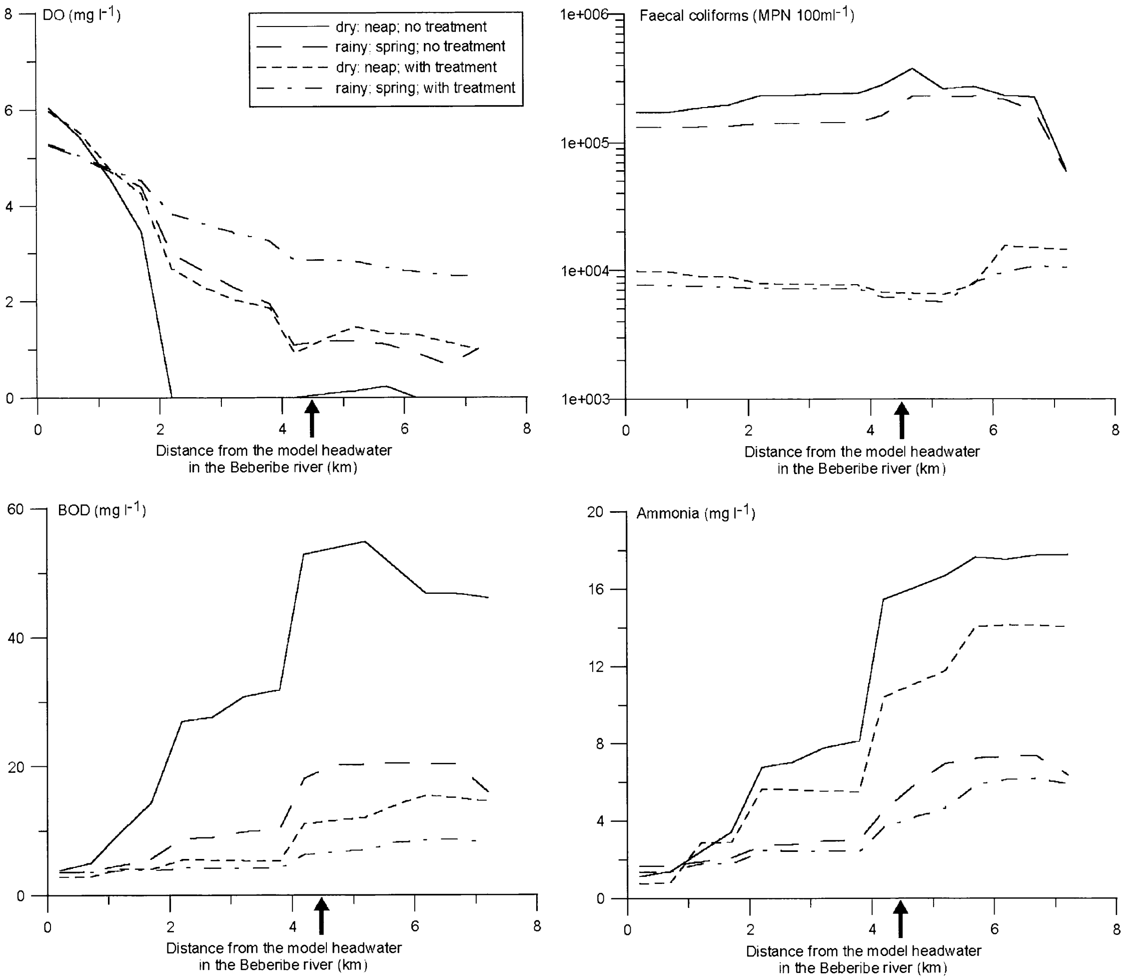

Figure 5. Dissolved oxygen (DO), faecal coliforms, biochemical oxygen demand (BOD) and ammonia model results, for the lowest (dry period and neap tide) and highest (rainy period and spring) dilution conditions, considering the 2020 expected population with and without sewage treatment. To simplify, the tributaries (Lavatripa and Morno streams) were excluded, showing here only the Beberibe river 8 km section. Arrows mark the tidal limit (4.5 km from the model headwater).

to the organic matter removal. This removal improves the DO minimum values from anoxic conditions to 2.0 mg l^{-1} in the lower 5 km of the Beberibe river during the dry season. For the rainy period, mean DO concentration at the same estuarine sector increases from 3.0 mg l^{-1} to 3.8 mg l^{-1} once the sewage plants become operational. Model results for ammonia, nitrate and phosphate did not show significant reductions because nutrient removal is not undertaken during primary sewage treatment. Ammonia and phosphate maximum values dropped from 17.5 mg l^{-1} to 14.0 mg l^{-1} and from 3.9 mg l^{-1} to 2.5 mg l^{-1}, respectively, during dry summer conditions. On high river flow simulations the ammonia maximum concentration decreased from 7.6 mg l^{-1} to 6.1 mg l^{-1}, and the phosphate, from 1.8 mg l^{-1} to 1.3 mg l^{-1}. Nitrate results did not show any difference after the sewage treatment had been carried out. These slight differences of the nutrient concentrations may be caused by the following factors: (1) changes to the nutrient load pattern from widespread sources to the localised sewage plant outflows; (2) the slight increase in DO, which can modify nutrient mass balance due to the oxygen dependent kinetic processes considered in the models.

Disinfection of the sewage treatment leads to a decrease of one order of magnitude in the number of FC, from 10^5 to 10^4 MPN 100 ml^{-1}. However, improvement of the dissolved oxygen conditions was not so satisfactory as the BOD and FC concentrations. The C_f/C_{sat} ratio (Table 1) also shows a deficit in the oxygen reaeration compared to the oxygen concentration necessary to oxidise the organic matter loads. This oxygen deficit was present in the estuarine sector even under high river flow conditions and became more evident during the drier period, even reaching the upper river. These hypoxic conditions were still found after most of the sewage organic matter had been removed.

Discussion

The strong influence of the hydrodynamics on the water quality variables in the Beberibe system was expected, due to the differences observed in the averaged values of the water quality monitoring program dataset used in the calibration and verification phases. During summer months, when river flow is reduced because of lower precipitation, water quality deteriorates compared to the rainy winter months. Characteristic low flow, resulting in small sewage dilution capacity, high retention time of pollutants and large turnover time, leads to this critical situation in the water quality. Braga et al. (2000) described a similar situation along inner channels of the Baixada Santista estuarine system, SE Brazil, where the weak hydrodynamics causes a lower dilution of the nutrient loads. The authors also related the hypoxic conditions and degraded water quality to the longer residence time in urban artificial channels in the Santos city.

The modelling results showed that the sewage treatment proposed for the Beberibe basin will not be able to bring water quality up to acceptable levels, mainly during dry summer months. This situation could be more critical in El Niño event years, when local winter rainfall is drastically reduced. In respect to the Brazilian water quality legislation (CONAMA, 1986), the Beberibe system would have unsuitable water quality in 20 and 17 of the 28 modelled segments for low and high river flow respectively, after the full primary treatment has been applied including disinfection. Suitable water quality conditions are not achieved due to low concentrations of DO and high BOD. As Beberibe water temperature varies from 26°C to 29°C, the warmer summer water may impede a major reoxygenation of the water because of lower oxygen saturation concentration. This may be partially responsible for the poor oxygen improvements and lower organic matter degradation rates.

Taking the old adage of sanitary engineering 'the solution to pollution is dilution' from Clark (1997), the Beberibe system should actually not bear any sewage discharge. The modelling results shown here indicate that Beberibe system will not reach the water quality required by the legislation even after all the sewage treatment has been implemented, although proposed sewage treatment plans would improve water quality in the estuary and streams. Results indicated a large decrease in FC concentrations from disinfection of the sewage plant outflows, and reduction on the DOB, but only a slight increase in DO content, due to lower organic matter disposal. However, the key aspect of the governmental program is the enhancement of public health as a consequence of better sanitary conditions. By keeping the population away from the sewage by serving them with adequate sewerage facilities, it is expected that there will be a large decrease in occurrence of related diseases, such as cholera, typhoid and amebiasis, making a dramatic improvement in the quality of life of this population.

All the results shown here were presented to the State Environmental Agency, CPRH, and to the Water Supply and Wastewater Company, COMPESA, to give them a scientific basis for a further discussion about the efficiency of the sewage program and the consequent water quality improvement in the Beberibe basin. We believe this can provide a powerful tool for the policy-makers choosing the best alternative for tackling the sewage problem. Some recommendations were given to the State Environmental Agency, CPRH, to adapt the monitoring plan to the water quality modelling proposed, such as locating sampling stations in the vicinity of the sewage treatment plants outflow points. Inclusion of water quality sampling and river flow measurement stations at the headwater of Morno and Lavatripa streams would enhance confidence in the modelling results.

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E. Orive, M. Elliott & V.N. de Jonge (eds), Nutrients and Eutrophication in Estuaries and Coastal Waters.

The prediction of nutrients into estuaries and their subsequent behaviour: application to the Tamar and comparison with the Tweed, U.K.

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Key words: dissolved nutrients, catchment modelling, river modelling, estuarine modelling, Tamar Estuary, River Tamar, Tweed Estuary

Abstract

Some of the techniques used to model nitrogen (N) and phosphorus (P) discharges from a terrestrial catchment to an estuary are discussed and applied to the River Tamar and Tamar Estuary system in Southwest England, U.K. Data are presented for dissolved inorganic nutrient concentrations in the Tamar Estuary and compared with those from the contrasting, low turbidity and rapidly flushed Tweed Estuary in Northeast England. In the Tamar catchment, simulations showed that effluent nitrate loads for typical freshwater flows contributed less than 1% of the total N load. The effect of effluent inputs on ammonium loads was more significant (~10%). Cattle, sheep and permanent grassland dominated the N catchment export, with diffuse-source N export greatly dominating that due to point sources. Cattle, sheep, permanent grassland and cereal crops generated the greatest rates of diffuse-source P export. This reflected the higher rates of P fertiliser applications to arable land and the susceptibility of bare, arable land to P export in wetter winter months. N and P export to the Tamar Estuary from human sewage was insignificant. Non-conservative behaviour of phosphate was particularly marked in the Tamar Estuary. Silicate concentrations were slightly less than conservative levels, whereas nitrate was essentially conservative. The coastal sea acted as a sink for these terrestrially derived nutrients. A pronounced sag in dissolved oxygen that was associated with strong nitrite and ammonium peaks occurred in the turbidity maximum region of the Tamar Estuary. Nutrient behaviour within the Tweed was very different. The low turbidity and rapid flushing ensured that nutrients there were essentially conservative, so that flushing of nutrients to the coastal zone from the river occurred with little estuarine modification.

Introduction

The purpose of this paper is to bring together and present some of the techniques used to model nutrient discharges from a terrestrial catchment to an estuary and to discuss the behaviour of these nutrients within the estuary. The Tamar catchment, its river system and estuary are used for purposes of illustration (Fig. 1). Although this study is local (regional) in focus, the modelling methodology and concepts may be of value to nutrient budget studies at the global scale, which rely largely on river-based measurements of nutrient concentrations (Howarth et al., 1996). Once river nutrients have entered their estuaries, transformations may occur before they then enter the coastal sea. For example, Balls (1994) demonstrated a clear link between conservative mixing of dissolved nutrients (with respect to salinity) and flushing times for a range of Scottish east-coast estuaries. Because estuarine nutrient behaviour depends upon estuarine properties, this paper also presents a discussion of the similarities and differences exhibited by nutrients transported within two contrasting estuaries (the Tweed and Tamar).

Nutrient discharges to rivers have sometimes been held responsible for increased phytoplankton growth and toxic algae development in estuaries and coastal seas (e.g. Cadeé, 1989; Riegman et al., 1992; Boynton et al., 1995). In addition, nitrogen compounds may exert a significant oxygen demand in estuaries through microbially mediated transformations (e.g. Owens, 1986; Grabemann et al., 1990). Organic nitrogen can be hydrolyzed to ammonia and additional, catchment derived ammonia may be input to estuaries via tributary rivers and wastewater discharges. Ammonia utilizes dissolved oxygen during nitrification to produce nitrate, via nitrite (e.g. Owens, 1986). In conditions of low dissolved oxygen, nitrate may be reduced to enable oxidation of organic carbon to proceed (e.g. Gameson, 1981). Estuarine waters are often enriched in phosphate from urban and industrial wastewater discharges and from land runoff (e.g. Hager & Schemmel, 1996).

On a global scale, it has long been recognised that human activity has greatly altered the nitrogen cycle in the atmosphere, in aquatic systems and on land (Berner & Berner, 1987). Global fixation of atmospheric N_2 for fertilizer, in combustion of fossil fuels, and by leguminous crops currently exceeds that by all natural sources, and changes in land use cause large additional amounts of nitrogen to be released from long-term vegetation and soil reservoirs (Vitousek, 1994). The environmental concerns are not restricted to estuarine and coastal eutrophication. They include also acidification of freshwater lakes and streams (e.g. Murdoch & Stoddard, 1992), forest decline (Schulze, 1989), climate change (Vitousek & Matson, 1993), and shifts in community structure (e.g. Bowman et al., 1995) and in ecosystem function (e.g. McNulty et al., 1994).

Methods

Modelling approach in the catchment

Several models, including: *P-EXPERT*, *ECM (Export Coefficient Modelling)* and *INCA-N* have been developed to budget and quantify diffuse- and point-source nutrient transfers from land to watercourses in order to predict pollutant loads both in-stream and at the tidal limits of estuaries.

The INCA model

The Integrated Nitrogen in Catchments model (*INCA*) simulates the hydrology, plant–soil system and stream–water, inorganic nitrogen (N) dynamics within a river catchment (Whitehead et al., 1999). The river catchment is sub-divided into reaches and, because land-use is one of the most important factors affecting N dynamics, the land-use within each reach is further sub-divided into six land-use groups. Spatial variations in land management (fertiliser application and timing), effluent discharge, N deposition and precipitation are taken into account. Detailed mass-balance equations are used to describe the production and transformation of NO_3-N and NH_4-N in each of the land-use types within the plant–soil system and also within the stream. By calibrating the model to observed stream water NO_3-N and NH_4-N concentrations, it is possible to use the equations to estimate fluxes (kg N ha^{-1} a^{-1}) for different processes in the plant-soil system, including mineralisation, immobilisation, nitrification and denitrification (Whitehead et al., 1999).

The ECM model

Export Coefficient Modelling (*ECM*; Johnes, 1996; Johnes et al., 1996) calculates the total load of N or phosphorus (P) arriving in a water body as the sum of the individual nutrient loads exported from each separate nutrient source in the catchment. The export coefficients are functions of the rate of nutrient input to each source and the nutrient export potential of each land-use type, livestock variety, or people. *ECM* also takes into account atmospheric deposition inputs:

$$L = \sum_{i=1}^{n} E_i[A_i(I_i)] + p. \tag{1}$$

L is loss of nutrients in Equation (1). E_i is export coefficient for nutrient source i. A_i is area of catchment occupied by land-use type i, or numbers of livestock of type i, or number of people. I_i is input of nutrients due to source i, and p is input of nutrients from precipitation.

The export coefficients, E_i, express the rate at which N or P is exported from each land-use type in the catchment. For animals in the catchment, the export coefficients express the proportion of wastes voided by them that will subsequently be exported from stock houses and grazing land to the drainage network. The amount of time each livestock type spends in stock housing, the proportion of the wastes voided which are subsequently collected and applied to the land in the catchment, and the loss of nitrogen through ammonia volatilisation during storage of manure are taken into account. For human wastes, the export coefficients reflect the use of phosphate-rich detergents and dietary factors in the local population.

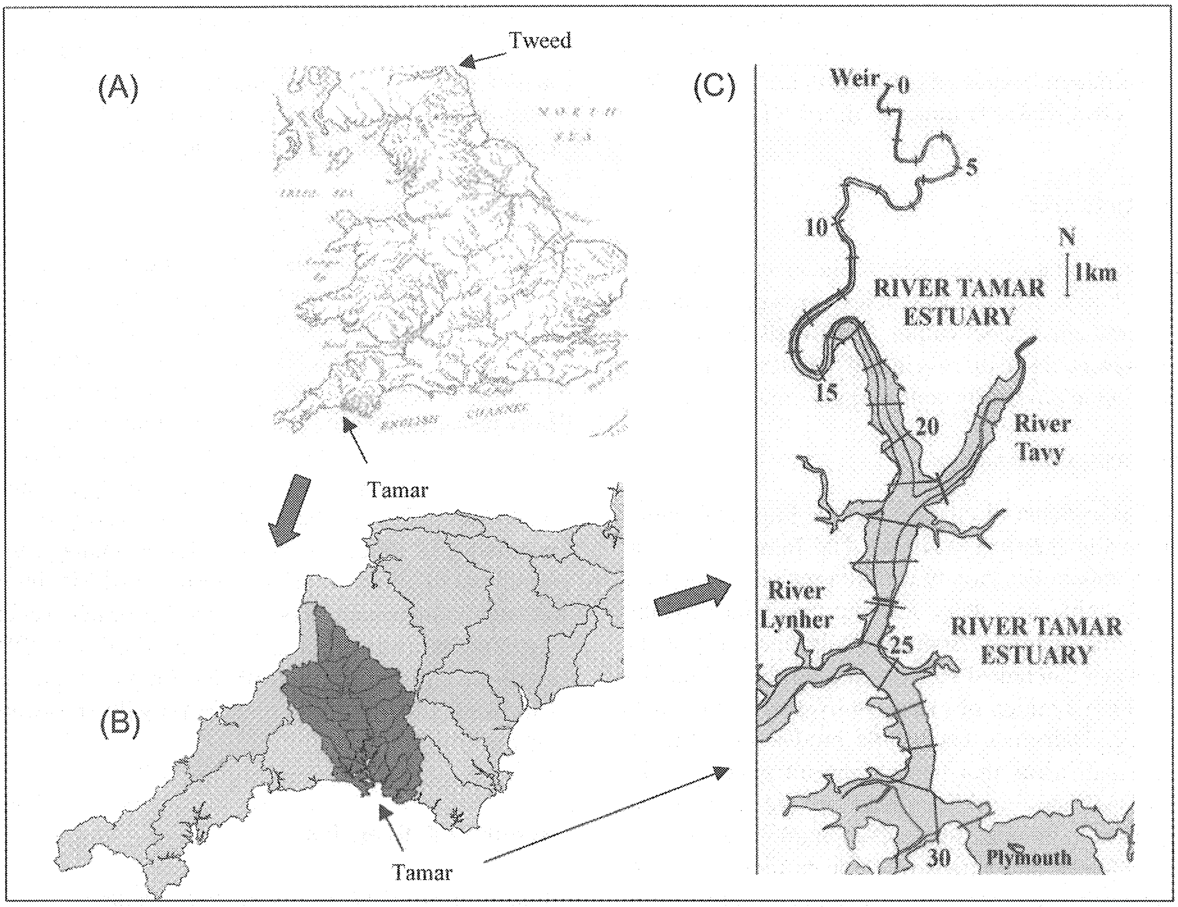

Figure 1. Catchment of the River Tamar and Tamar Estuary, SW England, U.K. (A), England and Wales; (B), Tamar catchment, SW England; (C), Tamar Estuary between the tidal limit at Gunnislake Weir and the English Channel at Plymouth. Along-axis distances are marked-off in 1-km segments.

The P-EXPERT model

The model *P-EXPERT* has been developed to provide a means of combining general estimates of total P loss from different soils and land-uses with site-specific information on erosion risk, as determined by soil texture and slope (Fraser, 2000). The model system provides estimated P transfer coefficients (PTCs) from agricultural sources along separate hydrological flow pathways. This technique is appropriate for use at the catchment scale.

Estuarine modelling

Complex time-stepping models of estuarine nutrient transport for the Humber Estuary are discussed by Tappin et al. (This Volume) and can equally well be applied to the Tamar. For purposes of illustration, however, the simplest, tidally averaged estuarine nutrient model is considered here of a steady-state estuary in which there is a constant freshwater inflow rate (e.g. Uncles, 1979). In this case, if conservative nutrient inputs are considered and these enter only in the freshwater at the head (tidal limit) then nutrient concentration will correlate linearly with the fraction of fresh water that constitutes a sample of estuarine waters. Alternately, if the only nutrient inputs considered are conservative and they enter in the coastal waters at the mouth, then the nutrient concentration will correlate linearly with the fraction of coastal seawater that constitutes the estuarine waters. Mid-estuarine nutrient inputs will generate mid-estuarine peaks in nutrient concentration. For example, an additional, mid-estuarine source of nitrate may result from nitrification due to bacteria that are associated with suspended sediment in the high turbidity zone of estuaries such as the Tamar and the Firth of Forth

(Owens, 1986; Balls, 1994). Other, non-conservative *in-situ* processes at work are de-nitrification and bacterial remineralization (e.g. Helder et al., 1983; Helder & De Vries, 1983; Trimmer et al., 1998).

The study areas

The main area of focus here is the catchment of the River Tamar and Tamar Estuary. However, nutrient measurements made within the Tweed Estuary are also presented and discussed and compared with those made in the physically contrasting Tamar Estuary.

The Tamar Catchment

The River Tamar discharges into the English Channel via the Tamar Estuary (Fig. 1). The Tamar's freshwater catchment lies largely within a region of intensive mixed arable and dairy farming, underlain by impermeable bedrock. Approximately one third of the freshwater catchment consists of moorland fringe, lying within a region of extensive livestock and upland farming. Dairy and beef cattle production, together with small areas that have been cultivated for fodder crops and winter stock feed, are the predominant land-uses in the intensive, mixed arable and dairy regions. Sheep and extensive cattle farming are more common in the extensive livestock and upland farming regions of the catchment. The human population is scattered throughout the catchment in small villages and isolated farmsteads.

The Tamar's river systems are well developed, with extensive flood plains that are used primarily for agriculture. However, water quality in the River Tamar has been adversely affected by agriculture. High rainfall and heavy, slowly permeable soils restrict the grazing season for cattle to a period between April and November. Grazing beyond this period results in soil poaching and structural degradation of the soil surface, which can lead to modification of its hydrological properties.

The Tamar Estuary

The Tamar is a partially mixed estuary that is 31 km long from its tidal limit at Gunnislake Weir to its mouth at the city of Plymouth (Fig. 1). It has mean neap and mean spring tidal ranges at its mouth of 2.2 and 4.7 m, respectively. Tidal current speeds can exceed 1 m s^{-1} in the upper reaches during spring tides and 0.5 m s^{-1} in the lower reaches. Its long-term, minimum, mean and maximum freshwater inflows are 3, 34 and 290 m^3 s^{-1}. Suspended particulate matter (SPM) levels are high in the upper reaches at spring tides (typically 50–1000 mg l^{-1}) and vary predominantly with the spring-neap cycle.

The Tweed Estuary

The River Tweed and its tributaries drain a sparsely populated, mainly agricultural area on the eastern borders of England and Scotland (Fig. 1). The river and estuary are considered to possess high water quality. The tidal limit is about 13 km from the mouth at Berwick-upon-Tweed. Tides at the mouth are semi-diurnal with mean spring and neap ranges of 4.1 and 2.5 m, respectively. The long-term, monthly-averaged freshwater inflow to the lower reaches varies from a maximum of about 140 m^3 s^{-1} during January, to a minimum of about 30 m^3 s^{-1} during July. Maximum and minimum instantaneous inflows, recorded over the last 30 years, were 1800 m^3 s^{-1} and 8 m^3 s^{-1}. SPM levels are fairly small (typically 2–30 mg l^{-1}) and vary with wave activity at the mouth and with freshwater inflows.

Results and discussion

Application of the INCA model to the Tamar Catchment

The non-tidal river Tamar was divided into 12 reaches within the *INCA* model, which covered a predominantly grazed-pasture catchment of 924 km^2. The calibration year was selected as 1997 because rainfall was similar to the long-term mean and an acceptable amount of observed data was available. Observed nutrient data were available for one main-channel site within the River Tamar, so that mean nitrogen concentrations were also included for calibration.

Except for the large freshwater flows that occurred during February 1997, flows simulated by *INCA* matched the observed data very closely in each of the 12 reaches (shown for Gunnislake Bridge, immediately up-river of the Weir, Fig. 1, on the upper panel of Fig. 2). This discrepancy was due to a gap in the observed rainfall data. Nitrate was simulated reasonably well (central panel, Fig. 2) and the daily-averaged concentrations matched the general trend of observed, monthly data. The ammonium simulation appeared to be much poorer (bottom panel, Fig. 2). However, most of the observed values were at (or near)

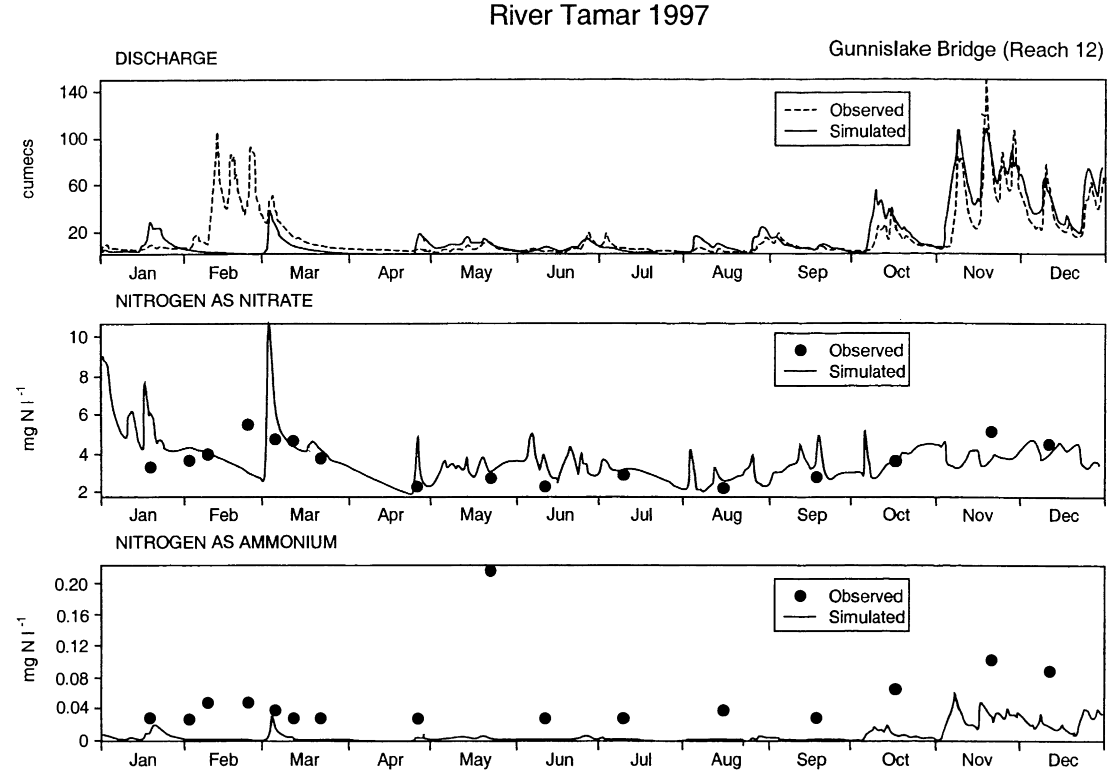

Figure 2. Calibration data for the *INCA* model, showing freshwater flows (top panel), nitrate (central panel) and ammonium (bottom panel) at Gunnislake Bridge, immediately up-river of Gunnislake Weir, during 1997.

the ammonium detection limit. Otherwise the general trend, if not the magnitude, was satisfactory. The ammonium value of 0.22 mg N l^{-1} during May was either erroneous or the result of an isolated pollution event in the river.

Input data for the validation simulation year of 1996 showed that this was an average year in terms of rainfall, with large rainfall events in the winter and autumn, storms at the beginning and end of the summer period, and occasional precipitation over the summer. The simulated freshwater flows matched the observed flows well (shown for Gunnislake Bridge, immediately up-river of the Weir, Fig. 1, on the upper panel of Fig. 3) although there was an overestimation at the end of the summer period, again due to inadequate input data for the *INCA* model. The nitrogen dynamics were reasonably well simulated. Observed trends and magnitudes of the nitrate concentrations were successfully reproduced (middle panel of Fig. 3). Ammonium concentrations were again close to the limit of detection (lower panel of Fig. 3).

Simulated data for the Tamar catchment indicate that effluent inputs did not make a significant contribution to the total nitrate loads during typical freshwater flows. Effluent nitrate loads during 1996–1999 contributed less than 1% of the total catchment load. The effect of effluent inputs on ammonium loads was more significant and effluents contributed around 10% of total catchment ammonium loads during 1996, 1998 and 1999. The ammonium contribution was more significant in 1997 due to lower freshwater flows than in the other simulated years. The simulated, average N export rate from the Tamar's catchment for the 4-year period 1996 through 1999 was 37 kg N ha^{-1} a^{-1}.

Application of export coefficient modelling to the Tamar Catchment

Export coefficients (E_i in Equation (1)) that were developed and validated for similar catchments elsewhere were applied to the Tamar's catchment. The N export rate from parishes (sub-areas of catchment) in 1991, estimated by export coefficient modelling (Fig. 4A), was dominated by export from cattle, sheep and permanent grassland in the catchment. The diffuse N export rate dominated the overall rate of nitrogen

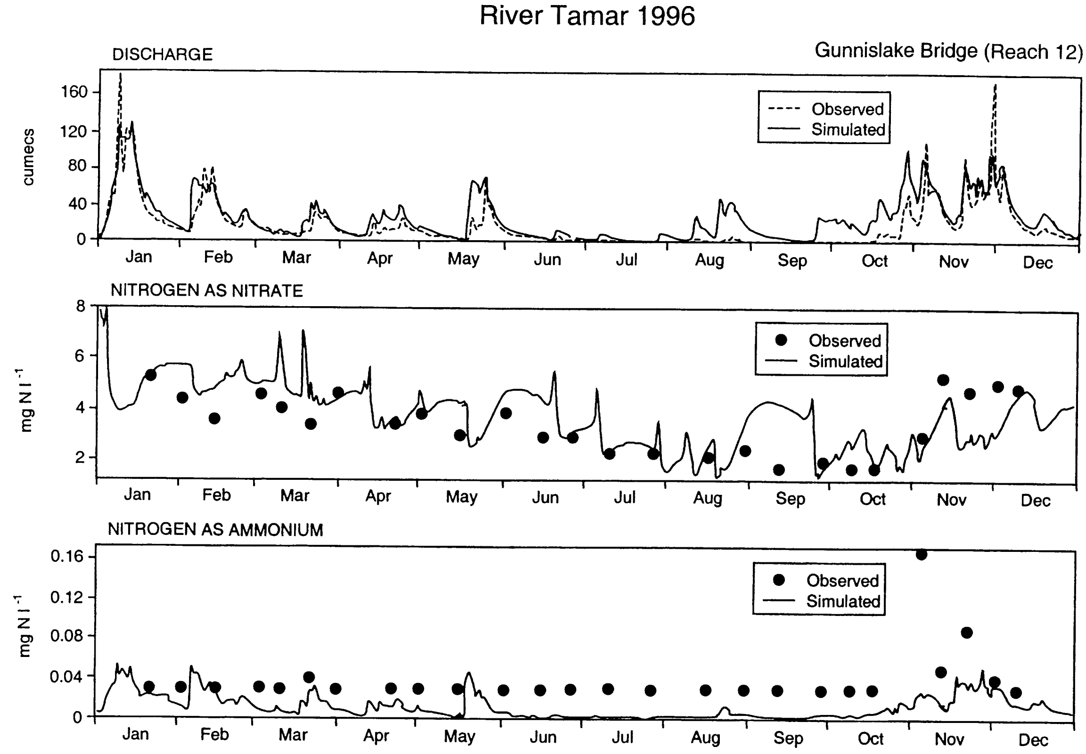

Figure 3. Validation run for the *INCA* model, showing freshwater flows (top panel), nitrate (central panel) and ammonium (bottom panel) at Gunnislake Bridge, immediately up-river of Gunnislake Weir, during 1996.

exported in 1991, with an average rate of 38 kg N ha^{-1} a^{-1} exported from diffuse sources, compared with 0.9 kg N ha^{-1} a^{-1} from point sources.

Cattle, sheep, permanent grassland and cereal crops generated the greatest rates of diffuse-source P export (Fig. 4B). This reflected the higher rates of fertiliser P applications to arable land and the susceptibility of bare arable land to P export in wetter winter months. Other important factors were the significance of livestock wastes as sources of P export from high stocking-density grazing land, and the high proportion of intensive grazing land in this catchment. The average rate of diffuse P exported in 1991 was 1.4 kg P ha^{-1} a^{-1}, while that from point sources was 0.2 kg P ha^{-1} a^{-1}.

Application of P-EXPERT Modelling to the Tamar Catchment

Hydrologically effective rainfall (defined here as excess winter rainfall) mostly occurs in the winter or field-capacity season and is a primary mechanism for P transfer. Movement of soil particles due to soil erosion and subsequent overland soil flows on susceptible, arable fields can transfer large amounts of P. Relative flows along various soil-association pathways were estimated as percentage values of hydrologically effective rainfall, using potential hydrological flow models. Using the best environmental data available for 1997, the total P transfer from the Tamar's catchment was computed as 1.4 kg P ha^{-1} a^{-1}. There is a close agreement between P exports derived from *ECM* and *P-EXPERT* (1.6 and 1.4 kg P ha^{-1} a^{-1}). These contrasting approaches use different driving parameters and it is encouraging that this measure of agreement exists.

The global context

The Tamar export of 37–39 kg N ha^{-1} a^{-1} during 1991 can be placed into a global context. Average exports into the North Atlantic from the NW coast of Europe, the watersheds surrounding the North Sea and the NE coast of the U.S.A. amount to 13, 15 and 11 kg N ha^{-1} a^{-1}, respectively (Howarth et al., 1996). Whilst these European N exports represent the greatest inputs per unit area and per unit time of all the coastal areas bordering the North Atlantic

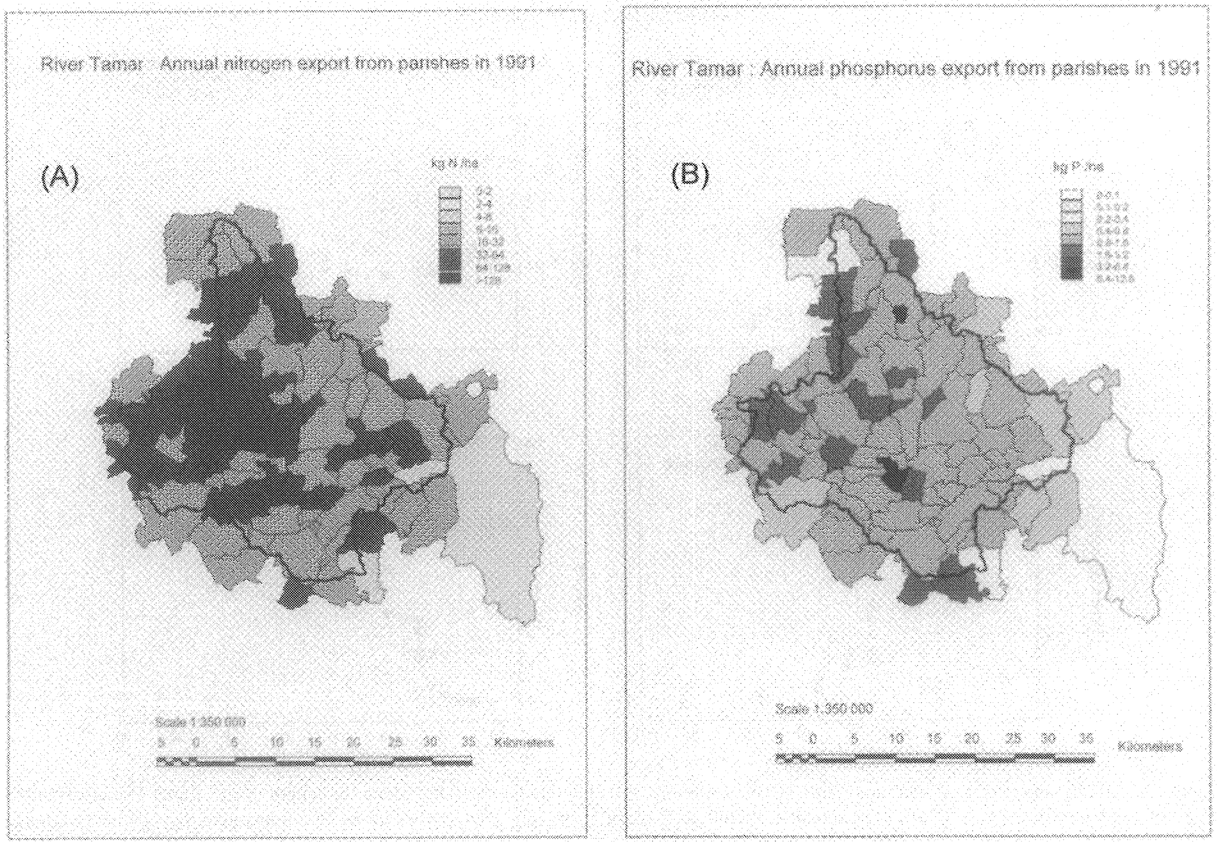

Figure 4. Simulated nitrogen (kg N ha^{-1}, (A)) and phosphorus (kg P ha^{-1}, (B)) nutrients exported from parishes (small areas of local government) of the Tamar Catchment during 1991, computed using Export Coefficient Modelling.

(Howarth et al., 1996), they are significantly less than that estimated for the agriculturally-dominated Tamar catchment. However, the Tamar export represents only 0.03% of the estimated 13 Tg a^{-1} of N input to the North Atlantic from the land (Howarth et al., 1996). Similarly, the P export of 1.4–1.6 kg P ha^{-1} a^{-1} during 1991 can be compared with 0.8, 1.2 and 1.4 kg P ha^{-1} a^{-1} from the NW coast of Europe, the watersheds surrounding the North Sea and the NE coast of the U.S.A., respectively (Howarth et al., 1996). The Tamar P exports represent only 0.007% of the estimated 2.3 Tg a^{-1} of P input to the North Atlantic from the land (Howarth et al., 1996).

Salinity, SPM and nutrients in the Tamar Estuary

The terrestrial catchment models supplied data on water flows and nutrient concentrations to the estuary via the river inputs. Once in the estuary, a nutrient's behaviour depended not only on the nutrient species, but also on the estuary's flushing rate, turbidity and other physical and biochemical variables (e.g. Morris et al., 1981; Nixon et al., 1996; Trimmer et al., 1998).

Recent measurements demonstrated that non-conservative behaviour was particularly marked for phosphate (16 July 1999, Fig. 5B). Phosphate exhibited a sharp spatial decrease in the tidal, freshwater reach, up-estuary of the freshwater-saltwater interface (FSI, located about 5 km from the tidal limit on Fig. 5D). The concentration of a conservative, hypothetical nutrient that possessed the same river and coastal levels as phosphate was much greater in the low-salinity upper reaches (Fig. 5B). Silicate concentrations were slightly less than conservative levels (Fig. 5A) whereas nitrate was essentially conservative (Fig. 5C). Concentrations of all three nutrient species were much smaller in the coastal waters than in the river waters, so that the coastal sea acted as a sink for terrestrially derived nutrients.

In-situ modifications of silicate (Fig. 6A) and phosphate (Fig. 6B) during their transport through the Tamar manifested themselves as non-linear scat-

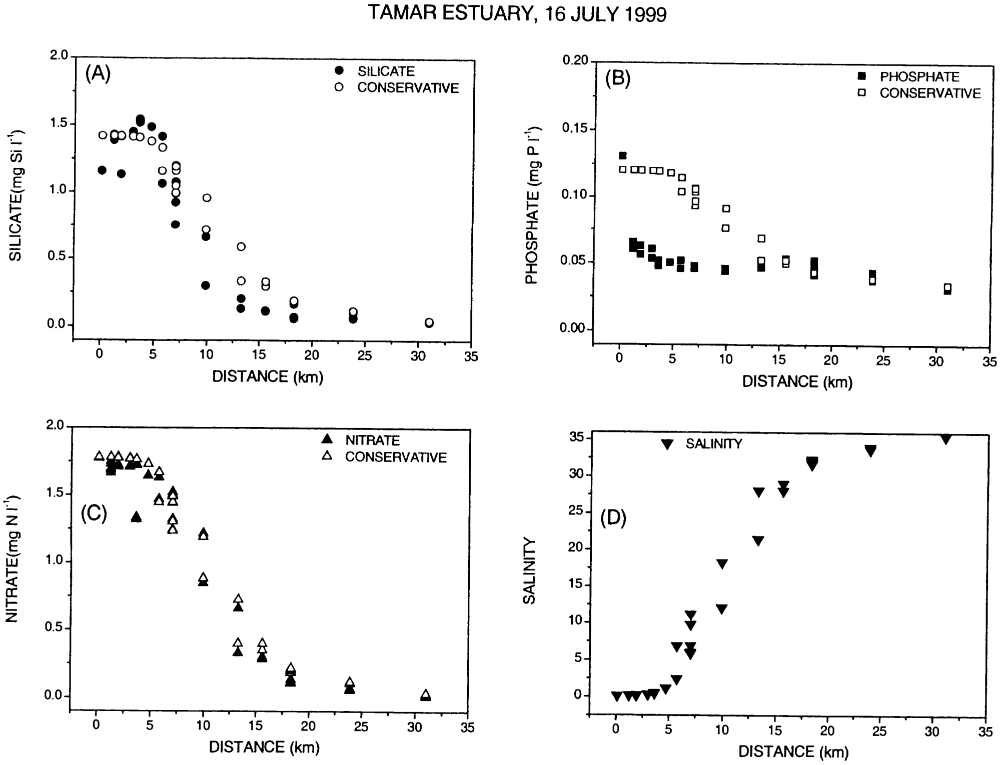

Figure 5. Salinity and dissolved inorganic nutrient concentrations (filled symbols) measured along the length of the Tamar Estuary from its tidal limit at Gunnislake Weir (0 km) to its mouth at Plymouth (31 km) on 16 July, 1999. Distributions of conservative, hypothetical nutrients that have the same river and coastal concentrations as the measured nutrients are shown by open symbols. (A), Silicate; (B), Phosphate; (C), Nitrate; (D), Salinity.

ter plots of concentration against salinity. The non-conservative behaviour of phosphate up-estuary of the FSI, and to a much lesser extent silicate, was a consequence of the strong turbidity maximum (TM) of fine-grained SPM that occurs in this region of the Tamar.

The differences in nutrient behaviour that can occur between estuaries due to their different physical properties are illustrated here using measurements of dissolved inorganic nitrate and nitrite in the estuaries of the Tamar and the Tweed (Fig. 1). The tidal reaches of the Tweed and Tamar exhibit very different behaviour, despite the fact that both have large tides (tidal ranges >4 m at mean springs) and strong freshwater runoff. The principal differences are geographical size, turbidity and flushing (or residence) times. Flushing times are of the order of one ebb tide for the Tweed (Uncles & Stephens, 1996) and two weeks for the Tamar (Uncles & Stephens, 1990). Freshwater spates (freshets), therefore, have an immediate and dramatic effect on the Tweed, leading to scouring of the bed and flushing of fine sediment and other materials to the coastal zone, where they are lost to the estuary. An inability to store significant amounts of fine sediment leads to a much lower turbidity in the Tweed than in the Tamar (2–30 compared with 10–1000 mg l^{-1} of SPM).

Unlike the Tamar, the distributions of silicate and phosphate concentrations were essentially linear when plotted against salinity (Fig. 6C, D), which indicates approximately conservative behaviour and relatively insignificant modifications during the rapid transport of these nutrients through the Tweed Estuary. In contrast to the nitrogen and silicon species, dissolved phosphate is affected by adsorption onto suspended sediment (Balls, 1994). This may explain why phosphate concentrations in the Tamar decreased so strongly in the high-turbidity waters down-estuary from the tidal limit at Gunnislake (Fig. 6B). Globally, it is known that intertidal sediments have a greater potential for P burial than for N burial (Billen et al., 1991; Howarth et al., 1995).

Although there was large scatter, the distributions of nitrate and nitrite concentrations in the Tweed were

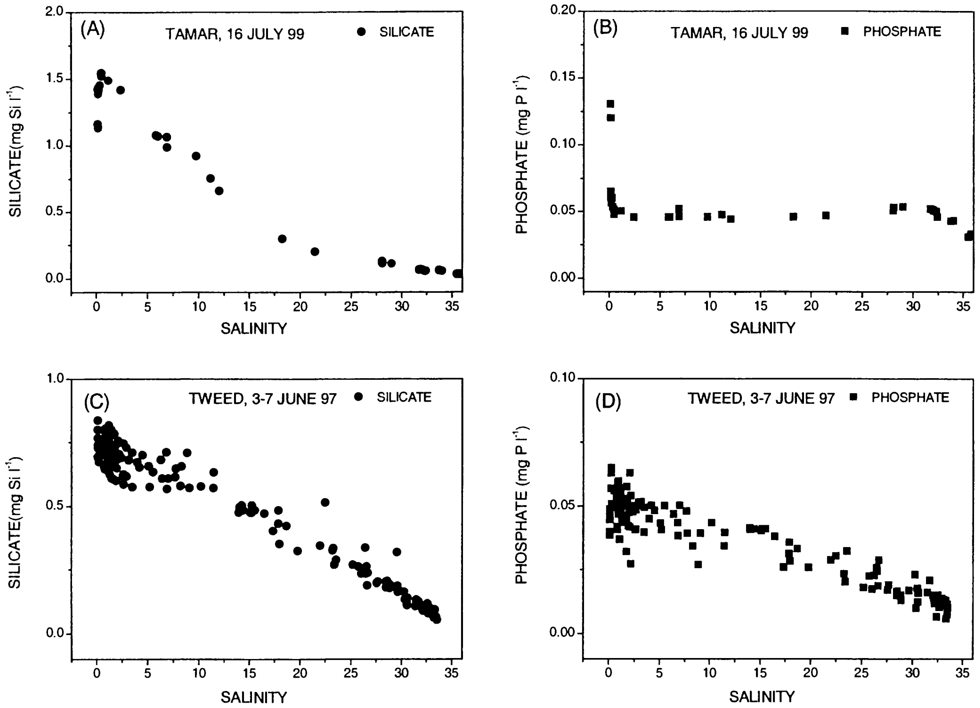

Figure 6. Scatter plots of dissolved inorganic nutrient concentrations against salinity measured in the Tamar (16 July, 1999) and Tweed (3–7 June, 1997) Estuaries. (A), Tamar silicate; (B), Tamar phosphate; (C), Tweed silicate; (D), Tweed phosphate.

approximately linear when plotted against salinity (Fig. 7A). Therefore, these nutrients were conservative to a first approximation, i.e. they were derived mainly from the freshwater inputs and were not significantly used-up or added-to during their transport through the rapidly flushed Tweed. Turbidity was low and SPM concentrations were less than about 30 mg l^{-1} (Fig. 7B) and much less than that towards the tidal limit (0 km). Higher turbidity occurred near the mouth (10 km) because of wave-induced suspension of bed sediments in the coastal zone and subsequent transport of coastal sediments into the Tweed during the flooding tide. Dissolved oxygen (DO) levels were essentially saturated throughout the estuary (Fig. 7B). This was a consequence of high DO in the freshwater inputs and coastal sea, together with rapid flushing of waters from the Tweed. In addition, there are only relatively unimportant biochemical oxygen demand (BOD) inputs to the estuary and low SPM concentrations (negligible uptake of DO by bacteria on the sediments; Trimmer et al., 1998).

Nitrate was essentially conservative within the Tamar (Fig. 7C) and nitrite behaved in an approximately conservative fashion for almost all the salinity range. Nitrite, however, was highly non-conservative at very low salinity (<2, Fig. 7C), near, and up-estuary of, the FSI. The measured freshwater concentrations of nitrite (0.008 mg N l^{-1}) and ammonia (0.03 mg N l^{-1}) were very low, which is consistent with output from the *INCA* model for the River Tamar (Figs 2 and 3). These freshwater data were interpolated from measurements in the non-tidal river at Gunnislake on 29 June 1999 and 26 July 1999 (the lower circles surrounding the nitrite and ammonia data on Fig. 7C). The interpolated river data for nitrate (2.4 mg N l^{-1}) was somewhat higher than that measured in the estuary on 16 July 1999, just down-estuary of the weir at Gunnislake (Fig. 7C). The pronounced peaks in nitrite and ammonia that occurred near the FSI subsequently determined the levels of these nutrients throughout the lower estuary. Therefore, unlike the Tweed, levels of estuarine nitrite (and ammonium) were determined by *in-situ* estuarine processes (e.g. Trimmer, 1998) rather than by typical freshwater levels, whereas freshwater levels largely determined nitrate concentrations.

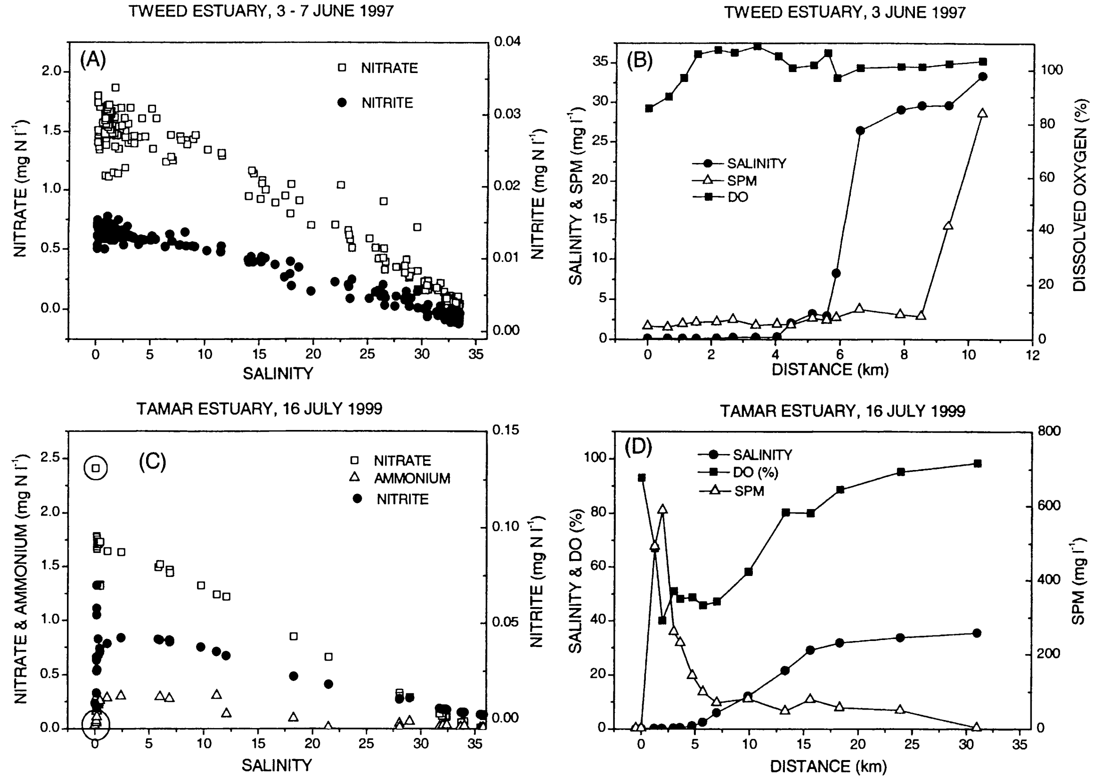

Figure 7. Salinity, dissolved oxygen (DO,% of saturation), SPM concentrations and dissolved inorganic nutrient concentrations in the Tweed (3–7 June, 1997) and Tamar (16 July, 1999) Estuaries. (A), Scatter plots of nitrate and nitrite versus salinity in the Tweed; (B), Surface salinity, SPM and DO as functions of distance from the tidal limit along the length of the Tweed Estuary on 3 June 1997; (C), Scatter plots of nitrate, ammonium and nitrite versus salinity in the Tamar – estimated river concentrations are circled; (D), Surface salinity, SPM and DO as functions of distance from the tidal limit along the length of the Tamar Estuary.

The *in-situ*, non-conservative nitrogen processes within the Tamar occurred in the vicinity of the TM. The TM was located immediately up-estuary of the FSI (about 2 km down-estuary of the weir at Gunnislake) and SPM concentrations within it were typically 600 mg l^{-1} (Fig. 7D). SPM concentrations were <50 mg l^{-1} much further down-estuary. The freshwater SPM concentration in the non-tidal River Tamar was approximately 2 mg l^{-1}. The high turbidity of the TM and the bacteria associated with its SPM and deposited sediments led to respiration of organic material and nitrification and de-nitrification processes, which together resulted in a depression of DO to about 40% of saturation (Fig. 7D). DO levels exceeded 90% in the freshwater River Tamar at Gunnislake and in the seaward reaches of the estuary (Fig. 7D). The DO minimum coincided with the nitrite and ammonium peaks and all were located a couple of km down-estuary from the TM peak. The reason for this displacement is that *in-situ* processes were taking place within the TM (rates were maximal there) at the same time as the affected waters were being flushed seawards.

Because nitrate distributions within the Tamar Estuary were essentially conservative, nitrate inputs from the river at Gunnislake largely determined concentrations of nitrate throughout the estuary. The nitrate concentrations in the River Tamar above Gunnislake Weir fluctuated greatly both throughout the year and over longer time-scales (Central panels of Figs 2 and 3). Highest nitrate concentrations tended to occur in winter, when freshwater flows were greatest, and the smallest concentrations in late summer and early autumn, when freshwater flows were also small (Central panels, Figs 2 and 3). The annual cycles of nitrate and freshwater flow show that nitrate lagged the flow by about 10–20 days, so that the annual nitrate peak occurred after the annual runoff peak. Rapid flushing of the Tamar occurred in winter during high flows, so that much of this nitrate was rapidly discharged to the coastal zone. In summer, flushing times were much longer and the lower nitrate inputs remained within the estuary for approximately two weeks (Uncles & Stephens, 1990) so that there was the possibility of *in-situ* uptake by algae at times of greater summer light exposure.

Concluding remarks

INCA is the only time-stepping model utilised here and was, therefore, the only model that could simulate seasonal and transient phenomena. It was used to compute river N concentrations and fluxes during 1996 through 1999. The *ECM* and *P-EXPERT* models essentially simulated long-term (annual) nutrient fluxes, given similarly long-term forcing functions. The close agreement between annual averaged N exports computed from *INCA* and *ECM* (37 and 39 kg N ha^{-1} a^{-1}, respectively) and between annual averaged P exports computed from *ECM* and *P-EXPERT* (1.6 and 1.4 kg P ha^{-1} a^{-1}, respectively) illustrates the effectiveness of these alternative approaches. Diffuse nutrient sources greatly dominated point sources and, on a global scale, Howarth et al. (1996) also found that non-point sources dominated the river fluxes of N to the coast in all regions of the North Atlantic.

This paper has considered the catchment scale and derived estimates of N and P exports using a limited selection of catchment parameters and variables. Regional to global scale models of N transport from land to ocean represent bulk N flux rates from large watersheds, regions, or continents, and similarly use a limited selection of parameters (Johnes & Butterfield, In Press). There are errors and uncertainties inherent in these existing regional to global scale models that propagate with increasing scale from small catchments to the regional scale. Increased understanding and improved modelling of catchment-to-estuary N (and P) fluxes can only improve our future ability to model at the global scale.

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Seasonal change in the proportion of bacterial and phytoplankton production along a salinity gradient in a shallow estuary

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Key words: bacterial production, specific growth rate, primary production, estuary

Abstract

We intended to evaluate the relative contribution of primary production *versus* allochthonous carbon in the production of bacterial biomass in a mesotrophic estuary. Different spatial and temporal ranges were observed in the values of bacterioplankton biomass (31–273 μg C l^{-1}) and production (0.1–16.0 μg C l^{-1} h^{-1}, 1.5–36.8 mg C m^{-2} h^{-1}) as well as in phytoplankton abundance (50–1700 μg C l^{-1}) and primary production (0.1–512.9 μg C l^{-1} h^{-1}, 1.5–512.9 mg C m^{-2} h^{-1}). Bacterial specific growth rate (0.10–1.68 d^{-1}) during the year did not fluctuate as much as phytoplankton specific growth rate (0.02–0.74 d^{-1}). Along the salinity gradient and towards the inner estuary, bacterio- and phytoplankton biomass and production increased steadily both in the warm and cold seasons. The maximum geographical increase observed in these variables was 12 times more for the bacterial community and 8 times more for the phytoplankton community. The warm to cold season ratios of the biological variables varied geographically and according to these variables. The increase at the warm season achieved its maximum in the biomass production, particularly in the marine zone and at high tide (20 and 112 times higher in bacterial and phytoplankton production, respectively). The seasonal variation in specific growth rate was most noticeable in phytoplankton, with seasonal ratios of 3–26. The bacterial community of the marine zone responded positively – generating seasonal ratios of 1–13 in bacterial specific growth rate – to the strong warm season increment in phytoplankton growth rate in this zone. In the brackish water zone where even during the warm season allochthonous carbon accounted for 41% (on average) of the bacterial carbon demand, the seasonal ratio of bacterial specific growth rate varied from about 1 to 2. During the warm season, an average of 21% of the primary production was potentially sufficient to support the whole bacterial production. During the cold months, however, the total primary production would be either required or even insufficient to support bacterial production. The estuary turned then into a mostly heterotrophic system. However, the calculated annual production of biomass by bacterio- and phytoplankton in the whole ecosystem showed that auto- and heterotrophic production was balanced in this estuary.

Introduction

The functioning and, in particular, the productivity of estuarine ecosystems is largely reflected in the levels of primary production and in the fate of autochthonous and allochthonous carbon. Changes in bacterial density and activity along trophic and salinity gradients affect differently the balance between carbon incorporation and carbon mineralisation in different parts of the estuary (Carlson & Ducklow, 1996; del Giorgio et al., 1997; del Giorgio & Cole, 1998). In the case of the mesotrophic ecosystem of Ria de Aveiro, most of the carbon which is uptaken by the water column bacteria is incorporated into biomass (about 90% for glucose) (Cunha et al., 2000). This is not the common conclusion derived from studies in other marine ecosystems where the bacterial growth efficiency in eutrophicated waters plateaux near 50% (reviewed by del Giorgio & Cole, 1998).

It is recognised that primary production plays a fundamental role on bacterial activity (Cole et al., 1988; Ducklow & Carlson, 1992; Conan et al., 1999) and that other factors, namely exogenous carbon (Mann & Wetzel, 1996; Goosen et al., 1997; Cunha et al., 1999; Reitner et al., 1999) may affect the coupling of these activities in aquatic ecosystems. On the other hand, physical and other chemical factors such as temperature, salinity and the concentration of mineral nutrients (PO_4^{3-} and NO_3^-) are also involved in the modulation of the bacterioplankton metabolism (Tabor & Neihof, 1982; Shiah & Ducklow, 1994; Riegsbee et al., 1996; Cunha et al., 2000; Almeida et al., 2001).

It is also known that the heterotrophic bacterioplankton consumes a large, although variable, fraction of the primary production (up to 50%) in marine and freshwater ecosystems (Cole et al., 1988; Lignell, 1990; Ducklow & Carlson, 1992; Conan et al., 1999). The phytoplankton community, however, does not always supply the total amount of carbon required for the observed level of heterotrophic bacterial growth (Scavia & Laird, 1987). In this case other substrate sources have to be admitted (Shiah & Ducklow, 1994). In this study, we intended to evaluate the importance of primary production in bacterial growth in a shallow estuary during the warm and cold seasons.

Materials and methods

Description of study site

Ria de Aveiro (see Fig. 1) is a mesotrophic shallow estuarine system on the western coast of Portugal connected to the Atlantic by a narrow opening. It is subjected to considerable inputs of industrial and domestic discharges occurring mainly at the periphery.

The lagoon covers an area of 66–83 km^2, respectively, at low and high tide (Dias et al., 2000). It exchanges with the sea 89 mm^3 of water in tides of 1–3 m amplitude (Silva, 1994). Several rivers carry an average of 1.8 mm^3 of fresh water into the lagoon during a tidal cycle (Moreira et al., 1993). The Ria has a complex topography, with three channels spreading from the mouth towards the different streams forming a multi-estuarine ecosystem. Secondary branches originate from the three main channels to form a tight net of channels, islands and mudflats. In this study, we chose the Canal de Ílhavo, the smallest of the second-order channels, with a water volume of 2.8 mm^3 at low tide and 9.3 mm^3 at high tide. This channel offers the advantages of a straighter water circulation and the absence of meaningful agriculture activity along the banks that might affect irregularly its trophic condition.

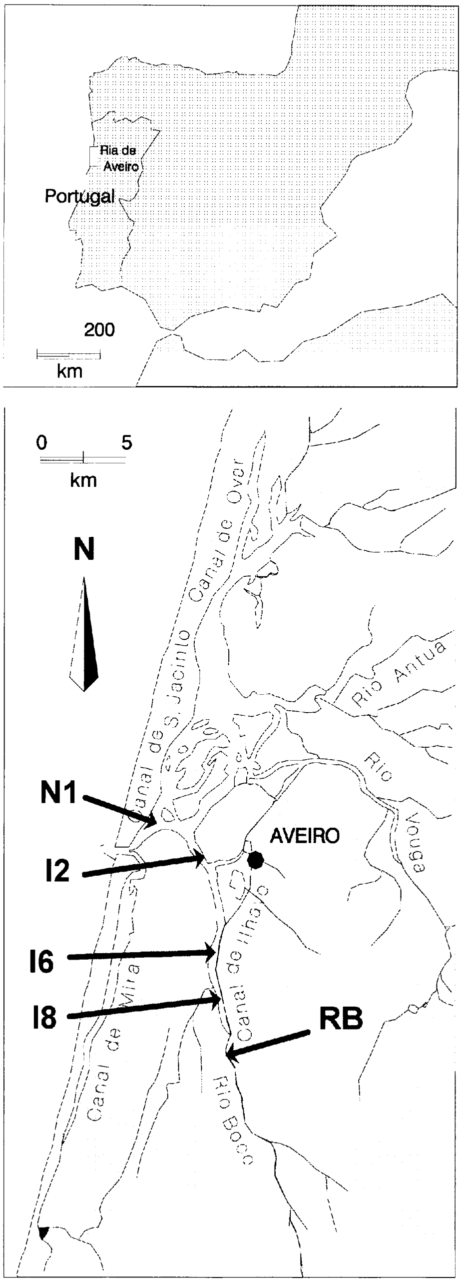

Figure 1. The estuarine system of Ria de Aveiro. Sampling stations are indicated by arrows along Canal de Ílhavo. Station N1 in Canal de Navegação represents the marine zone of the ecosystem. Stations I2, I6 and I8 represent the brackish water section of the salinity gradient along Canal de Ílhavo. Rio Boco (RB) is a river station although it is much affected by the variable strength of the tidal current.

The marine zone (MZ) of the ecosystem is represented by station N1 in Canal de Navegação. Stations I2, I6 and I8 represent the salinity gradient in the brackish water section (BZ). Rio Boco (RB) is the river station that is periodically affected by the variable strength of the tidal current.

Sampling

Water samples were collected along the longitudinal profile of salinity (see Fig. 1) in summer of 1997 (12th and 20th July). In the cold season, samples were collected only at two sampling sites (station N1, 10th December 1997 and 9th January 1998; station I6, 28th October and 7th November 1997). Surface water (0.2 m below surface) was collected at HT and LT at the centre of the channels.

The diel variation in bacterial production was studied in the marine zone during the warm season (16 and 17 of June 1997). In this case, surface water samples were collected at intervals of 1 h during a total of 24 h.

Water samples were transported to the laboratory and processed within 2–3 h after collection.

Physical and chemical variables

Water samples for nutrient (PO_4^{3-}, NO_3^-+NO_2^-) analysis were filtered through MSI acetate membranes with 0.45 μm pore size. PO_4^{3-} was determined by the molibdate method and NO_2^- plus NO_3^- were quantified by the sulfanilic acid method after reduction of NO_3^- to NO_2^- in a cadmium column. The analyses were performed in a Segmented Flow Injection Automatic Analyser Alliance Instrument – Evolution II (Abreu & Duarte, 1997). The concentration of suspended solids was determined after filtration of triplicate 0.5 l water aliquots through pre-weighted and pre-combusted Whatman GF/C filters. The filters were dried at 60 °C for 24 h and suspended solids (seston) calculated as the increase in dry weight. The organic matter fraction in the seston (POM) was determined as the loss of weight after 4 h combustion at 525 °C (Parsons et al., 1989). Particulate organic carbon (POC) was assumed to be 50% of POM (Rodier, 1996). The depth of the water column was measured with a Sonar probe (Hondex PS-7 LCD Digital Sounder). The turbidity of the water column was assessed as Secchi depth values (SD). Temperature and salinity values were read in a WTW LF 196 Conductivity Meter. Dissolved oxygen concentration, expressed as percentage of saturation, was determined with a WTW OXI 96 oxygen meter equipped with a WTW BR 190 stirrer.

Total bacterial number (TBN) and bacterial biomass (BB)

Bacterial cells were counted by epifluorescence microscopy with a Leitz Laborlux K microscope equipped with a I 2/3 filter for blue light. The samples were fixed with 2% formaldehyde (final concentration), filtered through black polycarbonate membranes of 0.2 μm (Poretics) and stained with 0.03% acridine orange (Hobbie et al., 1977). At least 200 cells or 20 microscope fields were counted in each of three replicate preparations. Bacterial biovolume was determined after measurement of 50 cells in each of triplicate samples. Cell volume was converted to cell carbon after assuming a conversion factor of 350 fg C μm^{-3} (Bjornsen, 1986).

Bacterial production (BP)

Bacterial production was determined in 10 ml triplicate sub-samples plus a control that was immediately fixed by addition of formaldehyde (2% final concentration). The samples were incubated in the presence of a saturating concentration (30 nM) of ^{3}H-leucine (Amersham, specific activity of 58–92 Ci $mmol^{-1}$). Incubation was carried out for 1 h at *in situ* temperature. After incubation, samples were fixed with formaldehyde. Protein precipitation was performed through the addition of 1 ml 20% ice-cold TCA followed by 15 min incubation in ice. These samples were filtered through 0.2 μm polycarbonate membranes (Poretics), rinsed twice with 2 ml of 5% ice-cold TCA and 5 ml of 90% ice-cold ethanol (Bligh et al., 1995). After standing for 3 days in scintillation cocktail (Universol) the radioactivity was read in a liquid scintillation counter (Beckman LS 6000 IC). Bacterial production was calculated on a per litre basis ($\mu g\ C\ l^{-1}\ h^{-1}$) and integrated to production per square meter ($mg\ C\ m^{-2}\ h^{-1}$) according to water depth. The total BP value for the ecosystem during the cold or the warm season was calculated from the averaged BP at the two tides (HT, LT) expressed on a per litre basis. These values were multiplied by 24 h not taking into account the eventual difference in production during night and day periods. The semester production (warm and cold seasons) in each zone resulted from diel production multiplied by 183 days and by the average volume of water in the respective zone – 32.1 mm^3 in the MZ and 88.6 mm^3 in the BZ – (Silva, 1994). The annual BP in the lagoon resulted from the sum of the two semester values.

Bacterial specific growth rate (BGR)

BGR was calculated as the ratio of bacterial production (BP), expressed in μg C l^{-1} h^{-1}, and bacterial biomass (BB) expressed in μg C l^{-1}. BGR per day respected a 24 h period.

Chlorophyll a (CHLO)

The concentration of chlorophyll *a* was determined fluorimetrically (Yentsch & Menzel, 1963) after filtration of 0.5 l triplicate samples through Whatman GF/C filters and overnight extraction in 90% acetone, at 4 °C. Phytoplankton biomass was calculated from the concentration of chlorophyll *a* assuming a conversion factor of 50 (Eppley et al., 1977).

Primary production (PP)

Primary production was determined by the ^{14}C-bicarbonate uptake method (Steeman-Nielsen, 1952). Duplicate samples (50 ml) were incubated with 8 μCi ^{14}C-bicarbonate (Amersham, specific activity of 50–60 mCi mmol^{-1}) at simulated *in situ* light conditions adjusted with neutral light filters to 100, 50, 25, 12,5, 1 and 0,1% of incident light intensity. After 4 h of incubation, 10 ml triplicate samples were filtered through a 0.2 μm pore-size membranes (Poretics polycarbonate membranes). The filters were treated with fuming HCl for 15 min and placed in picovials containing 5 ml of scintillation cocktail (Universol). The radioactivity was read immediately in a Beckman LS 6000 IC scintillation counter. Secchi depth was used for the calculation of the depths corresponding to 50, 25, 10, 1 and 0.1% of incident light intensity. Secchi depth was multiplied by an empirical factor, 0.40, 0.81, 1.35, 2.70 and 5.40, respectively, for 50, 25, 10, 1 and 0.1% of incident light intensity (K. Gocke, pers. com.). The integrated value, expressed in mg C m^{-2} h^{-1}, was calculated taking into account the production and the thickness of each photic layer. PP was then averaged according to the total volume of the water column (μg C l^{-1} h^{-1}). The total PP in the cold and warm seasons and the annual PP in the lagoon was calculated as described above for bacterial production but taking into account a 12 h period of insulation.

Phytoplankton specific growth rate (PGR)

The capacity of the autotrophic organisms to fix inorganic carbon was calculated as the ratio between PP expressed in μg C l^{-1} h^{-1} and the phytoplankton biomass expressed in μg C l^{-1} (Parsons et al., 1984). Specific growth rate per day was calculated multiplying that ratio by the average period of insulation (12 h).

Results

Physical and chemical variables

A clear longitudinal gradient of salinity was observed between station N1, in the marine zone, (33.5–35.6 PSU) and Rio Boco (4.5–7.3 PSU) (as shown in Table 1). In winter, the salinity values were 6–8 PSU lower when compared to summer values. Water temperature in the warm season varied between 16.6 and 26.6 °C increasing upstream in daylight. In the cold season, the temperature declined to 11.2–18.5 °C, not as much as usual for winter. The level of oxygen saturation in the water column fluctuated from 76 to 116%. POC ranged from 5.5 to 7.0 mg l^{-1} in the warm season, and from 6.7 to 9.5 mg l^{-1} in winter. The longitudinal profile of water depth varied from 0.5 to 7.7 m during the warm season and from 1.8 to 13.0 m in the cold months. Secchi depth was greater in the marine zone, ranging along the salinity gradient from 0.5 to 4.5 m in the warm season and from 0.8 to 2.0 m in winter. The concentration of NO_2^- plus NO_3^- in the warm season showed a longitudinal pattern of variation characterised by an enormous peak (111.6–204.6 μM) in the inner estuary. This peak did not reach the mid estuary and only a small variation in this concentration (0.9–5.5 μM) could be noticed from the mid-estuary to the mouth. The variation in the concentration of PO_4^{3-} (0.3–3.3 μM) was not so clearly patterned, but the lowest concentrations were always found at the outer estuary and the peaks at the mid-estuary (as shown in Table 1). The nutrients levels were usually higher at low tide throughout the longitudinal profile producing LT/HT ratios of 2.3 for NO_2^- plus NO_3^- and 4.4 for PO_4^{3-}. In winter, the nutrient levels in Ria de Aveiro, are usually up to 20 times higher for NO_2^- plus NO_3^- and up to 2 times higher for PO_4^{3-} (T. Vinhas, M Valença, unpublished data).

Biological variables

Bacterial biomass (BB) varied from 45 to 273 μg C l^{-1} in the warm season (see Fig. 2) establishing a sharp pattern of enrichment from the outer to the mid-inner estuary. In the cold season, BB decreased, in

Table 1. Physical and chemical variables. Salinity (SAL), temperature (TEMP), percentage of dissolved oxygen (%SAT O_2), depth, Secchi depth (SD), particulate organic carbon (POC), dissolved PO_4^{3-} (PO_4^{3-}), dissolved NO_2^- plus NO_3^- (NO_2^-+NO_3^-) and chlorophyll *a* (CHLO) in the warm and cold seasons at high (HT) and low tide (LT)

		SAL (UPS)	TEMP (°C)	%SAT O_2	DEPTH	SD (m)	POC (mg l^{-1})	PO_4^{3-} (μM)	NO_3^-+NO_3^{3-} (μM)	CHLO (μg l^{-1})
Warm Season										
HT	N1	35.6	16.6	78	7.5	4.2	6.0	0.26	1.42	6.9
(12th July 1997)	I2	34.7	18.5	78	7.7	3.5	6.0	0.38	1.41	8.2
	I6	32.3	21.3	78	2.4	1.8	6.5	1.20	2.37	12.1
	I8	29.0	23.3	78	2.3	1.0	6.5	1.01	2.84	33.5
	RB	7.3	24.1	116	1.0	0.5	6.5	0.40	204.60	22.8
LT	N1	33.5	208	96	5.8	2.0	6.5	0.89	0.87	11.2
(20th July 1997)	I2	33.3	22.1	92	4.2	1.2	6.0	1.45	2.21	9.6
	I6	28.9	24.9	76	1.5	0.8	6.0	1.66	1.96	19.2
	I8	25.2	25.9	ND	2.0	0.7	7.0	2.14	5.53	18.6
	RB	4.5	26.6	ND	0.5	0.5	5.5	1.48	111.60	13.0
Cold Season										
HT (9th January 1998)	N1	33.1	14.9	90	13.0	8.8	9.5	ND	ND	1.6
(7th November 1997)	I6	27.2	16.0	85	3.0	8.4	7.8	ND	ND	5.0
LT (16th December 1997)	N1	21.8	11.2	95	12.3	95	8.6	ND	ND	0.9
(28th October 1997)	I6	19.2	18.6	51	1.8	51	6.7	ND	ND	7.5

ND, not determined.

Table 2. Warm to cold season ratios of bacterioplankton and phytoplankton variables measured at high (HT) and low tide (LT) in the marine (MZ) and brackish water zones (BZ). Bacterial biomass (BB), bacterial production (BP), bacterial specific growth rate (BGR), phytoplankton biomass (PB), phytoplankton production (PP) and phytoplankton specific growth rate (PGR)

Variable	MZ (Station N1)		BZ (Station I6)	
	HT	LT	HT	LT
BB	1.4	3.0	2.2	1.7
BP	20.0	2.1	2.0	3.2
BGR	13.3	1.0	1.0	1.9
PB	5.0	12.0	2.4	2.6
PP	112.3	34.7	20.7	8.6
PGR	26.3	2.6	9.3	5.7

general, by a factor of 2 (as shown in Table 2). BB was higher at low tide (LT) in the marine and brackish water zones and in both seasons. Phytoplankton biomass (PB) showed also a clear longitudinal pattern of variation with a maximum value of 1675 μg C l^{-1} at station I8, in summer and at high tide (HT). In the cold months, the levels of phytoplankton biomass decreased 8.5 times at station N1 and 2.5 at station I6 (as shown in Table 2). At station I6, in the brackish water zone, phytoplankton biomass was higher at LT (1.5 ×) in opposition to station N1, in the marine zone, where phytoplankton was 1.8 times more concentrated at HT. Bacterial production (BP) was more intense in the brackish water stations of the lagoon. In the warm season, BP ranged from 1.1 to 16.0 μg C l^{-1} h^{-1} (6.3–36.8 mg C m^{-2} h^{-1}) along the longitudinal profile (see Fig. 3). In a 24 h tidal cycle in the marine zone during the warm season, BP decreased to around 50% during the dark period when compared to the diel average (see Fig. 4). At noon, BP was around twice the diel BP average while at mid-night, in the same tidal phase, BP was 1.6 times lower than the diel average and 2.7 times lower than the noon value. Bacterial production in winter (0.1–6.0 μg C l^{-1} h^{-1} or 1.5–17.7 mg C m^{-2} h^{-1}) decreased 2 times at LT and 20 times at HT in the outer estuary (station N1) and within a range of 2–3 times in the mid estuary (station I6) as shown in Table 2. However, the pattern of BP variation with tide was not clear in any of the two stations.

Phytoplankton production (PP) in summer varied along the longitudinal profile from 13.6 to 512.9 μg C l^{-1} h^{-1} (60.1–512.9 mg C m^{-2} h^{-1}) increasing from the outer to the inner estuary (see Fig. 3). The sharpness of the increasing profile was attenuated when PP was integrated according to water column depth.

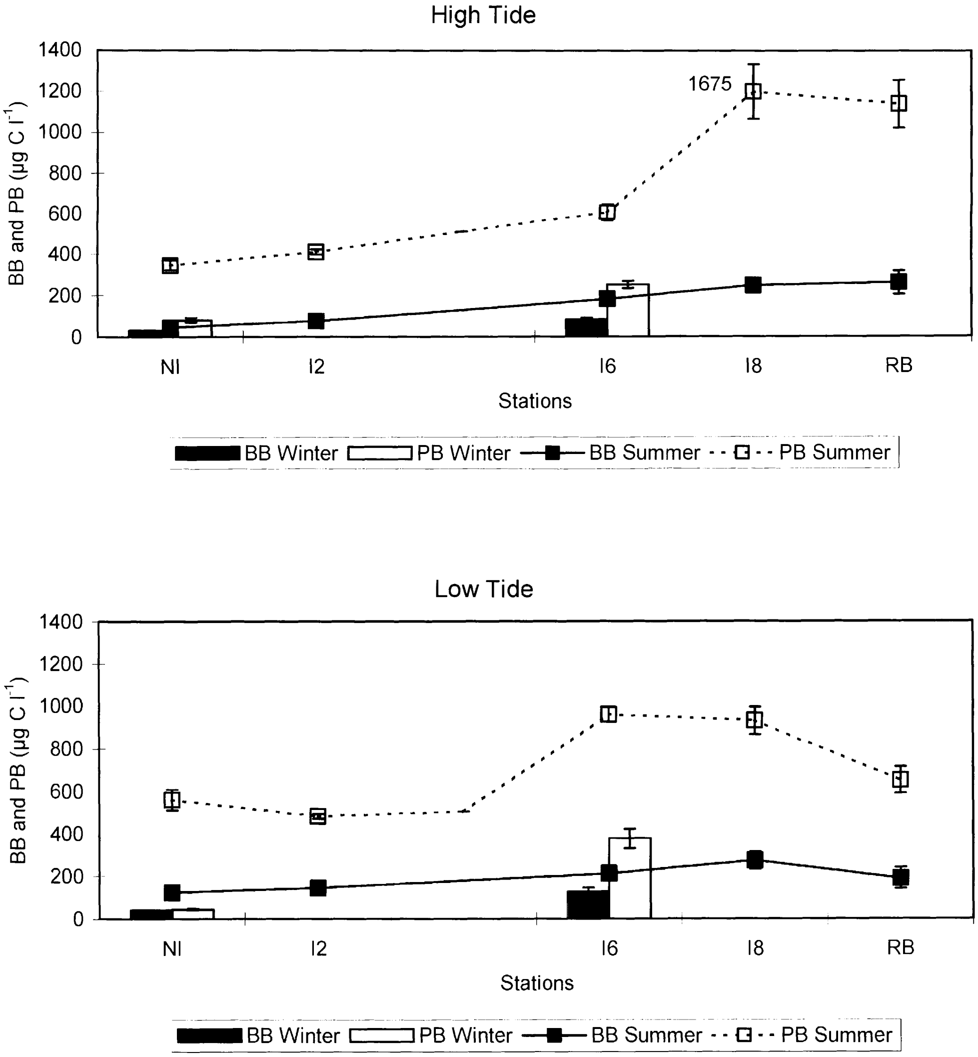

Figure 2. Bacterio- and phytoplankton biomass in the warm season (HT 12th July 1997, LT 20th July 1997) and cold season (Station N1, HT 9th January 1998 and LT 16th December 1997; Station I6, HT 7th November 1997 and LT 28th October 1997). Bacterial biomass (BB), phytoplankton biomass (PB).

In fact, although phytoplankton activity was lower in the marine zone, the greater water depth of this zone increased the integrated value. In the cold season, primary production varied from 0.11 to 3.3 μg C l^{-1} h^{-1} (1.5–6.0 mg C m^{-2} h^{-1}). Compared to summer values it decreased 35–112 times at station N1 and 9–21 at station I6 (as shown in Table 2). The maximum values of PP per unit of volume were generally observed at LT.

In the marine zone, BP expressed on an annual basis and on a per unit water volume was lower (9201 μg C l^{-1} y^{-1}) than PP (32 574 μg C l^{-1} y^{-1}). In the cold season, however, BP (1425 μg C l^{-1} h^{-1}) amounted to a higher value than PP (606 μg C l^{-1} h^{-1}). On the other hand, in the BZ, BP (67 953 μg C l^{-1} y^{-1}) and PP (72 966 μg C l^{-1} y^{-1}) were similar on an annual basis (as shown in Table 3). Considering the total water volume of the lagoon (119.8 mm^3 being 31.2 mm^3 in the MZ and 88.6 mm^3 in the BZ) (Silva, 1994), BP summed to 6309 ton C y^{-1} and PP 7482 ton C y^{-1} fractionated as shown in Table 3.

In the cold and warm seasons, bacterial specific growth rate (BGR) generally increased in less saline waters (up to 6 and 18 times in summer and winter, respectively), showing a range of 0.10–1.68 d^{-1} (see Fig. 5). In the marine zone, BGR at HT was 13 times

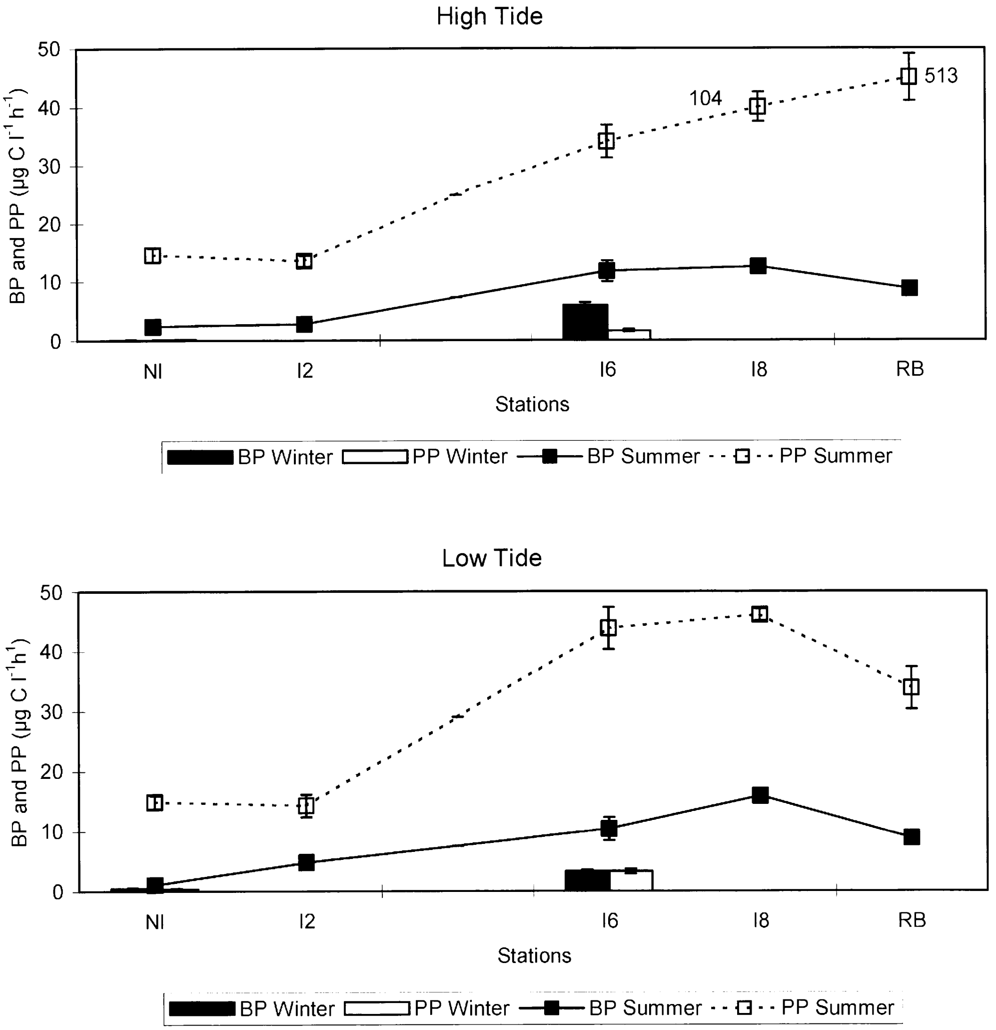

Figure 3. Bacterio- and phytoplankton production in the warm season (HT 12th July 1997, LT 20th July 1997) and cold season (Station N1, HT 9th January 1998 and LT 16th December 1997; Station I6, HT 7th November 1997 and LT 28th October 1997). Bacterial production (BP), phytoplankton production (PP).

lower in cold water than in summer but at LT, BGR was similar in both seasons. In the brackish water zone, BGR at HT was not very different in warm and cold weather, but at LT was about 2 times higher in the warm water. Phytoplankton specific growth rate ranged from 0.32 to 0.74 d^{-1} in the warm season and from 0.02 to 0.12 d^{-1} in winter (see Fig. 5). The PGR values generally increased from the marine to the brackish water zones (up to 2 and 4 times in summer and winter, respectively). In summer, PGR was 3 to 26 times higher than in winter. PGR maxima were observed at HT and coincided with PP maxima in both seasons.

In the warm season, BP/PP varied from 0.02 to 0.35 along the longitudinal profile of Canal de Ílhavo (see Fig. 6) with maxima at LT. The proportion of bacterioplankton biomass to phytoplankton biomass (BB/PB) in this season ranged from 0.13 to 0.30. Both ratios tended to increase from the outer- to the mid-inner estuary. In the cold season, however, BP/PP and BB/PB ratios increased to values of 1.00–3.61 and 0.34–0.91, respectively (see Fig. 6).

Discussion

Different seasonal, geographical and tidal conditions, involving temperature, salinity and nutrient supply, originated a wide range of bacterioplankton specific growth rates (0.14–1.68 d^{-1}). Warm weather en-

Table 3. Estimated values of the annual production of bacterio- and phytoplankton biomass in the marine zone (MZ), in the brackish water zone (BZ) and in the whole lagoon of Ria de Aveiro (MZ+BZ). Bacterial production (BP), phytoplankton production (PP)

		Warm season ton C 6 month^{-1} (μg C l^{-1} 6 month^{-1})	Cold season ton C 6 month^{-1} (μg C l^{-1} 6 month^{-1})	Total Annual ton C y^{-1} (μg C l^{-1} 6 y^{-1})
MZ	BP	243	45	288
(31.2 mm^3)		(7776)	(1425)	(9201)
	PP	999	18	1017
		(31968)	(606)	(32574)
BZ	BP	4248	1773	6021
(88.6 mm^3)		(47952)	(20001)	(67953)
	PP	5991	474	6465
		(67608)	(5358)	(72966)
MZ+BZ	BP	4491	1818	6309
(119.8 mm^3)		(55728)	(21426)	(77154)
	PP	6990	492	7482
		(99576)	(5964)	(105540)

mm^3, million cubic meter.

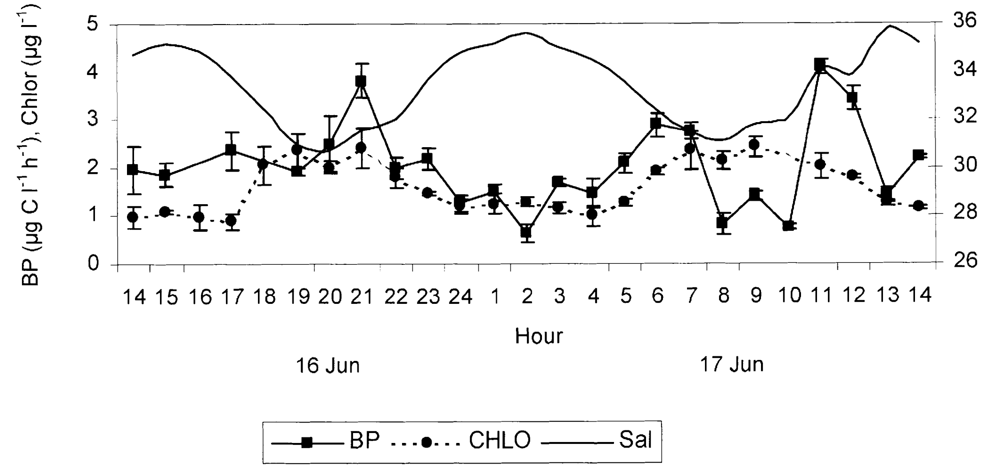

Figure 4. Diel fluctuation of bacterial production (BP) and chlorophyll *a* (CHLOR) in response to tidal current in the marine zone (warm season). Salinity (Sal).

hanced the specific bacterial growth rate by factors of up to 2 in brackish water and of up to 13 in marine water. The factor of increase in BGR fluctuated considerably during the tidal cycle. Previous laboratory experiments have demonstrated the strong response of bacterial activity to exposure to contrasting water properties (Cunha et al., 2001). Taking these results into consideration, the tidal fluctuation of BGR was interpreted as the result of poor growth of brackish water bacteria when transported to a high salinity environment as well as of the stimulation of marine water bacteria when transported to brackish water. The positive response of marine bacteria to brackish water is, however, counterbalanced to some extent by the low salinity. Preliminary results showed that the stimulation could be enhanced if salinity of brackish water was increased by the addition of NaCl (Almeida et al., 2001a). These facts resulted in an absence of a significant increase of BGR at LT in the marine zone as well as at HT in the brackish water zone during the warm season as might be expected. For this reason, the general increase in BGR from the mouth to the

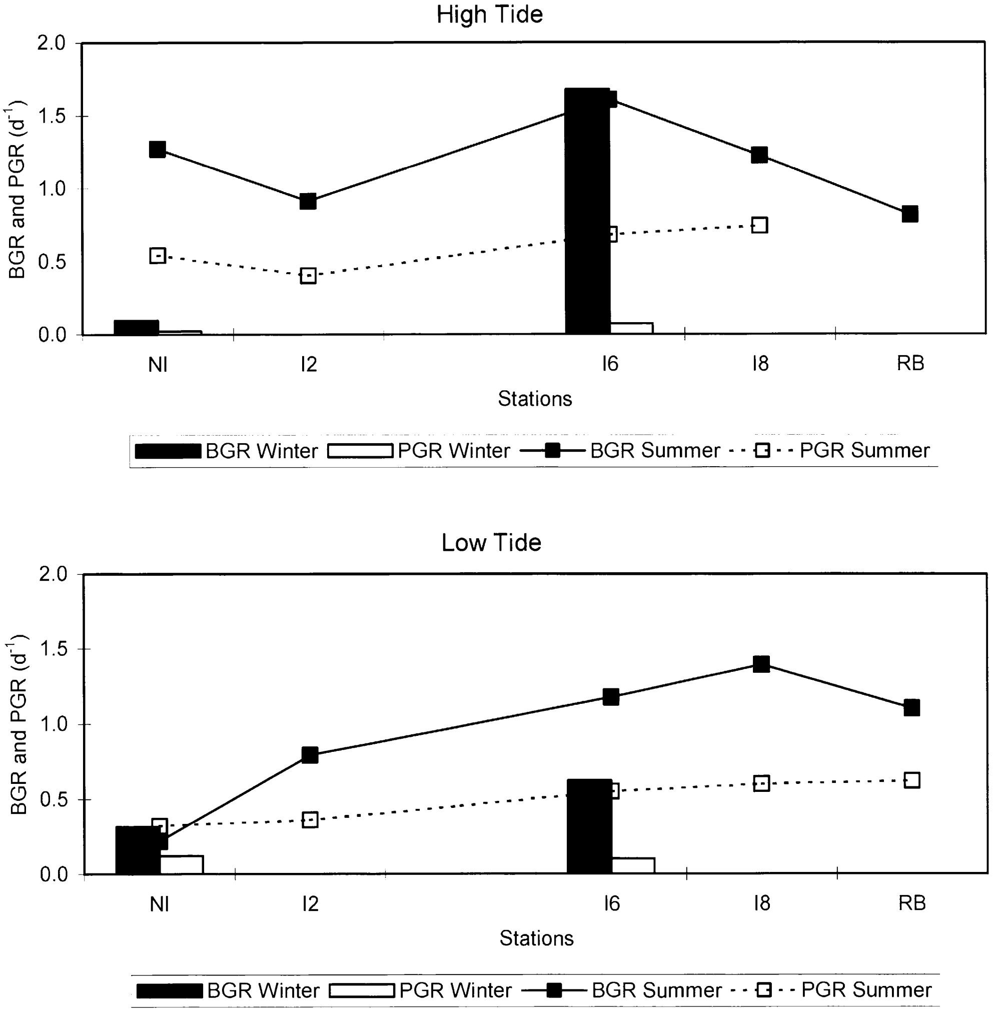

Figure 5. Bacterio- and phytoplankton specific growth rates in the warm season (HT 12th July 1997, LT 20th July 1997) and cold season (Station N1, HT 9th January 1998 and LT 16th December 1997; Station I6, HT 7th November 1997 and LT 28th October 1997). Bacterial specific growth rate (BGR), phytoplankton specific growth rate (PGR).

mid-inner estuary was clearer at LT. These facts taken together help to explain the observed spatial variation in bacterial production and reinforce our prior conclusion on the existence of bacterial communities with distinct salinity optima in the estuary (Almeida et al., 2001b). The seasonal and geographical variation of phytoplankton specific growth rate followed the same pattern as BGR but was affected by higher factors. Phytoplankton growth rate in the cold season decreased by factors of 3–26, probably due more to reduction of insulation than the small decrease in temperature, whereas BGR decreased by factors of 1–13. The greater (2–4 times) PGR values observed in the brackish water zone followed the increase in nutrients concentrations.

In the cold months and throughout the longitudinal profile of salinity, the bacterio- to phytoplankton biomass ratio was considerably high (0.34–0.91) suggesting that primary production may not be enough to support secondary production. Indeed, the production ratio (BP/PP) along the salinity gradient varied, during this season, from 1.00 to 3.61. We infer that in the cold season, the potential contribution of allochthonous carbon to bacterial biomass production was very high (close to 100%) in both zones of the lagoon.

In the warm season, the scenery in the lagoon changed. The bacterio- to phytoplankton biomass ratio along the longitudinal profile of salinity decreased to much less than 1 (0.13–0.30) both at HT and LT. These ratios suggest that bacterial production, in the

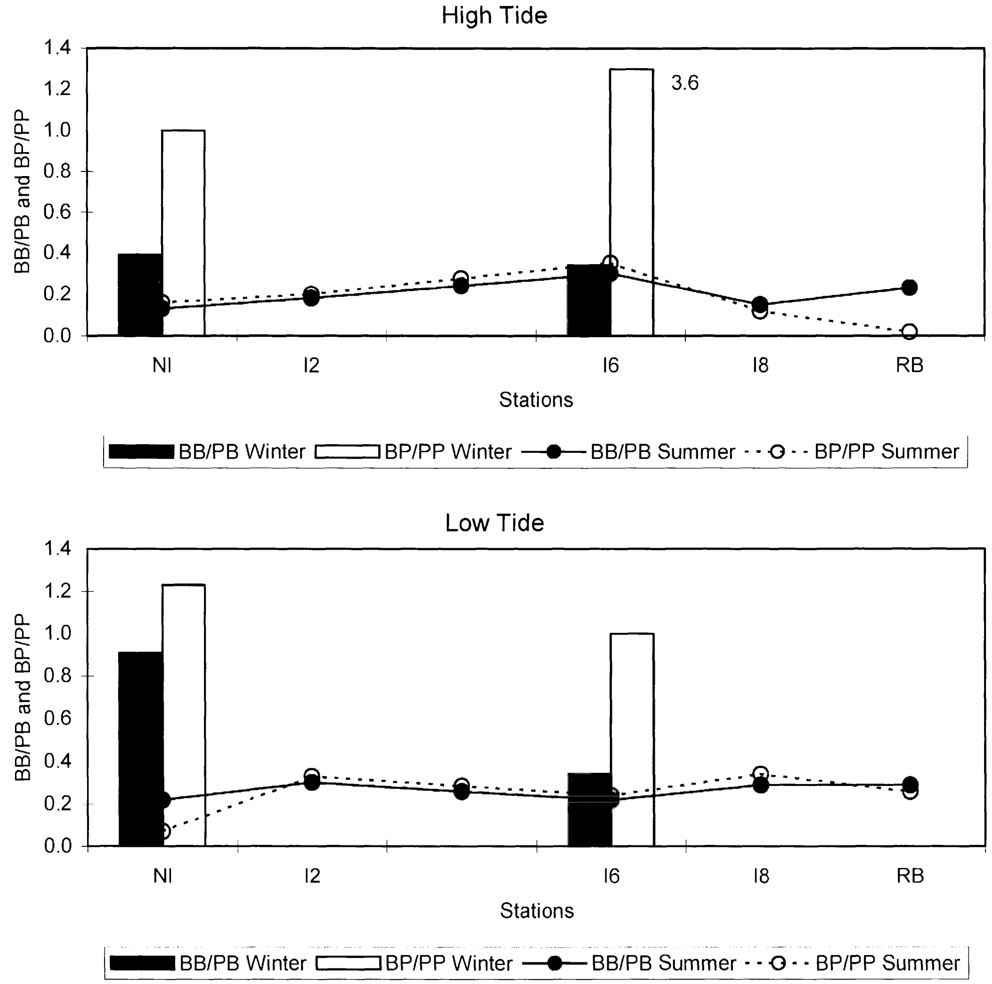

Figure 6. Bacterio- to phytoplankton ratios of biomass (BB/PB) and production (BP/PP) in the warm season (HT 12th July 1997, LT 20th July 1997) and cold season (Station N1, HT 9th January 1998 and LT 16th December 1997; Station I6, HT 7th November 1997 and LT 28th October 1997).

warm season, might be essentially supported by phytoplankton carbon. In fact, the results showed that BP/PP ranged from 0.02 to 0.35. The carbon incorporated in bacterial biomass at the different sampling stations corresponded, on average, to about 21% of primary production, an acceptable value as observed in other environments where phytoplankton could support the whole bacterial production (Cole et al., 1988; Kuosa & Kivi, 1989; Lignell, 1990; Fuks et al., 1994). Assuming that the contribution of primary production to bacterial production in winter is negligible, that the sources of allochthonous carbon are seasonally stable and that the water temperature did not vary drastically from one season to the other, the bacterial production in winter could be taken as a measure of allochthonous carbon inputs and used to more accurately estimate the contribution of phytoplankton to bacterial production in warm weather. The fraction of primary production diverted to the bacterial biomass production in the warm season ($BP_{summer} - BP_{winter}/PP_{summer}$) would decrease to 13%. These figures are within the range of the estimated 6–16% by Conan et al. (1999) for aquatic systems in general and of the experimental estimated 5–15% obtained by Fernandez et al. (1994) in the Mediterranean Sea. Neither it is far from the 20% value reported by Rheinheimer (1981) for the Kiel Bight. Furthermore, the observation of diel fluctuation in bacterial productivity, characterised by a decreased average during the dark period, suggests coupling between phytoplankton production and consumption by bacteria as reported by Gasol et al. (1998). Although during the warm season, primary production could be enough to support the whole bacterial production in the estuary, in the brackish water zone allochthonous carbon could contribute, nevertheless, to the production of a significant fraction of bacterial biomass. Taking into consideration the previous estimate of allochthonous organic carbon inputs, it is possible to estimate the contribution of primary production to bacterial production during the warm season by subtracting the fraction sustained by allochthonous sources from the total bacterial production in summer. The fraction of bacterial production sustained by primary production was estimated in this way as 59% of the total bacterial production in the brackish water zone and as 74% in the marine zone. This confirms the results of other authors who found that in mid-inner estuary the coupling between BP and PP was lower than in the outer estuary (Shiah & Duklow, 1995).

Conclusion

The specific growth rate of the bacterioplankton within the estuary was patterned according to space and season along the estuarine gradient. The presence of bacterial communities with distinct salinity optima, adapted to marine or brackish water conditions, seemed to affect the pattern at different tides.

The geographical, seasonal and diel variations in biomass production bring about the conclusion that on an annual basis and on a per water volume unit, the ratio bacterial production to primary production was less than 1 in the marine zone and almost 1 in the brackish water zone. This defines two broad subsystems where bacterioplankton and phytoplankton are in different balance in relation to biomass production.

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The planktonic food web structure of a temperate zone estuary, and its alteration due to eutrophication

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Abstract

Current conventional wisdom argues that human-induced excesses in nutrient loadings to estuaries often stimulate 'excess' algal production leading to hypoxia, via bacterial pathways, and subsequent reduced recruitment/survival of finfish and shellfish. Why wouldn't such elevated production stimulate more animal production, rather than less? In a three-year study of Long Island Sound, U.S.A., a multitude of variables were quantified along a west to east gradient, to address the above question via the hypothesis that different successes among planktonic species experiencing eutrophication alter planktonic food web structure away from traditional pathways to microbial loop dominated ones. Variables studied included: nutrient concentrations and ratios (i.e. NO_2, NO_3, NH_4, DON, PON, PO_4, Silicate, N/P and N/Si), phytoplankton, protozooplanktonic ciliate, zooplankton, heterotrophic nanoplankton (HNAN), photosynthetic nanoplankton (PNAN), size-fractionated chlorophyll, larval fish and bacterial concentrations and/or species composition, and bacterial growth rates (as frequency of dividing cells, FDC). Results indicated that although current nitrogen and other nutrient loadings into the estuary are much higher than past inputs (especially in western waters), the average concentration of dissolved inorganic nutrients is similar (though slightly higher) to past values. Relative proportioning among chemical species does vary from west to east, with NH_4 and dissolved organic nitrogen (DON) at times more prevalent in the west, especially in bottom waters. Excess loadings of nitrogen and other nutrients into the estuary are converted to elevated biomass of both small ($< 10\ \mu$m), and large ($>20\ \mu$m) phytoplankton in the west. Slightly enhanced bacterial densities and growth rates shadow the elevated chlorophyll levels, with distinctive Sound-wide seasonal patterns that follow not total chlorophyll, but rather PNAN concentrations. HNAN concentrations also are elevated in the west, and likely influence bacterial dynamics. Species composition of phytoplankton routinely differ west to east. Inorganic N/P are routinely low (i.e. below Redfield ratios), especially in the west, while total dissolved N/P (i.e. including DON) are similar among stations and typically are significantly higher than Redfield ratios. Associated with bacterial and $<10\ \mu$m chlorophyll enhancements to an elevated diversity of ciliate species in the west. Copepod biomass is extremely enhanced in the west, indicating that while stimulating the microbial loop, eutrophication is also enhancing the secondary production preferred by larval fish and gelatinous zooplankton. Larval fish diversity is down relative to the past, but shows little contemporaneous west/east variations. So, if adult fish populations are down, but larvae are not food limited, possibly toxicity, overfishing, and/or habitat destruction which prevent a healthy, normal system response to eutrophication are culpable. It is suggested that recipients of the excess copepod production are likely gelatinous zooplankton and benthic sediments, and that unused copepod 'excess' biomass likely significantly contributes to hypoxia.

New conventional wisdom: Excess nitrogen stimulates microbial loop and net phytoplankton biomass and production, which in turn stimulates microcrustasean biomass and production and fecal release, and both significantly fuel hypoxia and likely stimulate gelatinous zooplankton production.

Introduction

Current conventional wisdom regarding many naturally mesotrophic to eutrophic estuaries argues that excess, anthropogenically introduced, nitrogen stimulates phytoplankton production, which goes underutilized, settles to the bottom of the estuary, and leads to the development of hypoxia via bacterial decomposition pathways, with concommitant decreases in recruitment and survival of commercially important finfish and shellfish (EPA Report, 1994).

Why shouldn't fertilization of these types of estuaries, and associated increases in algal biomass, over multi-year time scales, stimulate secondary & higher production leading to enhancement of finfish production rather than to hypoxia (Capriulo et al., 1993, 1997)? It is known that elevated nitrogen inputs affect plankton dynamics (Murdoch et al., 1998) and generally favor net phytoplankton over nanoplankton (Margalef, 1978; Demers et al., 1986; Kiørboe, 1993). Such phytoplankton are the preferred food of microcrustacean zooplankton, which in turn are preferred food for many larval fish. So, initially one might expect differential, time dependent growth responses of phytoplankton *versus* zooplankton, to result in increased algal biomass fluxes to the sediments, with associated elevations in microbial activity. However, eventually, over appropriate time scales, larger zooplankton, and in turn fish should respond to cyclically consistent elevated food levels with higher recruitment success and therefore enhanced populations and biomass. If eutrophication results not in enhancement of net phytoplankton, macrozooplankton and fish, but rather in stimulation of the microbial loop, it might explain widescale scientific and anecdotal reports of deteriorating finfish and shellfish stocks (Capriulo et al., 1993, 1997; EPA Report, 1994).

To attempt to address this question, we set up the hypothesis that differential successes among planktonic species experiencing eutrophic conditions is altering planktonic food web structure in the western Long Island Sound away from 'traditional pathways', and towards microbial loop dominated pathways. This study addresses several levels of questions designed to test the above hypothesis in the near shore waters of Long Island Sound, to develop contemporary baseline data on the plankton food web structure and associated nutrient dynamics of nearshore Long Island Sound waters, to search for west to east eutrophication-related gradients in microbial loop dynamics, nutrient and chlorophyll concentrations, overall water column biomass, and species composition (e.g. of phytoplankton, protozooplankton and copepods) structure , and to relate any observed differences to macrozooplankton and larval fish population dynamics. The data is here synthesized into an updated version of Riley et al.'s work of the 1950s (Riley, 1955, 1956a,b; Riley et al., 1956, 1959; Riley & Conover, 1956, 1967), expanded to cover the microbial loop, a more complete nutrient balance sheet (i.e. NO_2, NO_3, NH_4, DON, PON, PO_4, Silicate), and determination of nutrient ratios. The primary goal of this research was to describe the nearshore planktonic food web structure of Long Island Sound, and to determine if it has been changed by eutrophication. Also, if such change is evidenced, to consider whether the changes have forced the system towards a primarily microbially dominated structure, and away from a more 'traditional' macrozooplankton and larval fish structure.

Eutrophication and the question of a changed ecosystem

Eutrophication in marine systems refers to natural or artificial additions of limiting nutrients, and to the ensuing changes (Rohlich, 1969). Nitrogen is considered the primary limiting nutrient in LIS and most other coastal marine systems (Riley & Conover, 1956; Harris, 1959; Ryther & Dunstan, 1971; Mann, 1982). The prime effect of nitrogen addition on phytoplankton is an increase in primary production, but often overlooked are concommittent and related alterations of species composition and cell size distribution (Lund, 1969; Ketchum, 1969; Uye, 1994; Murdoch et al., 1998; Agawin et al., 2000).

Successful recruitment, growth and production of finfish and shellfish in aquatic ecosystems depend heavily on food quality and therefore habitat quality (Cushing, 1975; Jones, 1976; Lasker, 1981). In particular, year classes are made or lost at the larval stages of development, with food size, quality, species composition and concentration present during larval critical growth periods determining survival rates (Lasker, 1981). For this reason, it is important to analyze and understand water column properties and dynamics that influence the structure of planktonic food webs leading to finfish and shellfish, in coastal waters and estuaries such as Long Island Sound. Anthropogenic inputs that disturb food web dynamics in Long Island Sound must therefore be identified and resultant changes in interactions understood before effective remediation can occur.

Carbon fixed by primary producers is transported along food webs through both 'copepod' and 'microbial' pathways, toward higher levels, but the proportions directed along each route depend in large part on species composition, prey quality and size (Walker & Peterson, 1991; Thingstad & Rassoulzadegan, 1999), as well as related water column chemistry and mixing dynamics. It is believed that pathways involving copepods are more beneficial to finfish populations (Greeve & Parsons, 1977; Capriulo, 1990; Capriulo et al., 1991).

The 'Microbial loop'

In recent years, it has become clear that the microbial loop (Pomeroy, 1974; Azam et al., 1983) is an integral component of most marine/estuarine food webs, and that microbial loop dynamics influence the abundance, diversity and production characteristics of fish and shellfish populations, ctenophores and crustaceans (Sherr et al., 1986; Capriulo, 1990; Capriulo et al., 1991). The term microbial loop is used to refer to planktonic (and epi) bacteria, cyanobacteria, heterotrophic (HNAN) and photosynthetic (PNAN, which includes mixotrophs) nanoplankton, dinoflagellates, ciliates and amoebae. These organisms interact intra- and inter-specifically in a number of complex ways, and are at the center of research interest and a paradigm shift with respect to our understanding of aquatic food web structure. Protistans have higher weight specific nutrient regeneration (Zeuthen, 1943; Johannes, 1964, 1965; Fenchel & Finlay, 1983; Goldman et al., 1985; Caron & Goldman, 1990) rates than do larger zooplankton (e.g. copepods) and thus dramatically influence water column chemistry (e.g. dissolved gases, nutrients, etc.) when they are present in significant concentrations (Capriulo et al., 1991; Thingstad & Rassoulzadegan, 1999). With respect to feeding, phagotrophic protists remove up to 100% of the daily bacterial production and up to 60% of the yearly primary production in different marine systems (Capriulo et al., 1991). Additionally, many protozooplankton and benthic protists can endure (and some even thrive under) exposure to 'poor water quality' (e.g. hypoxia) better than larger forms such as the metazoans, and thus will persist in 'polluted' habitats (Fenchel et al., 1990; Malvin & Wood, 1992). Despite this core of interest in the importance of the microbial loop to marine ecosystems, little research has been carried out in this area for Long Island Sound.

Microzooplankton in LIS have a community ingestion rate similar in magnitude to that of the copepod community (Capriulo & Carpenter, 1980, 1983). Microzooplankton are the main trophic link to copepods in systems where most photosynthesis is carried out by cells too small for copepods to effectively consume, e.g. coastal waters after the spring bloom (Sherr & Sherr, 1988). The microbial heterotroph sector of the food web consists of a network of bacteria and protozoa driven to a large extent by dissolved organic matter (DOM). The DOM pool is maintained via leakage of photosynthate through cell walls of algae (Larsson & Hagstrom, 1982), 'sloppy feeding' by copepods (Capriulo et al., 1988; Capriulo, 1990; Peduzzi, 1992), leaching from fecal pellets of copepods (Jumars et al., 1989), the decomposition of organic particles by bacteria using enzymatic hydrolysis (Banse, 1992), and sewage and riverine inputs to an estuary such as LIS. Releases of DOM account for 20–40% of mean primary production (Azam & Fuhrman, 1984; Hagstrøm, 1984). Bacteria feed on the DOM, and in turn are food for heterotrophic flagellates (HNANS) and ciliates (Gast, 1985). The HNANS are eaten by microzooplankton that copepods may eat (Azam et al., 1983; Ducklow, 1983; Andersen & Sorensen, 1986; McManus & Fuhrman, 1988; Capriulo, 1990; Capriulo et al., 1991; Dolan, 1991; Hoch & Kirchman, 1993). Heterotrophic microbes perform a vital function in the food web by efficiently regenerating the nitrogen in DOM to an inorganic form (Caron & Goldman, 1990; Capriulo, 1990; Thingstad & Rassoulzadegan, 1999). This function is particularly vital when limiting nutrients are all but undetectable in the spring and summer.

Bacteria

Naturally occurring planktobacterial concentrations in marine ecosystems typically range from 10^3 to 10^7

per ml, with estuaries and other coastal waters often supporting concentrations in the 10^6 per ml range (Fuhrman et al., 1980; Kirchman et al., 1982; Ducklow, 1983; Capriulo, 1990). Concentrations of bacteria in water proximal to sewage and/or storm water drainage sites often reach concentrations in the 10^9 per ml range in large part due to heightened coliform bacteria levels. It has been estimated that 10–50% of all primary production in coastal waters passes through the bacterioplankton with as much as half of that being incorporated into new bacterial biomass (Hagstrøm et al., 1979; Fuhrman & Azam, 1982). Many scientists now believe that bacteria are indispensable to the well being of many phytoplankton, which rely on various bacterial products (e.g. vitamins) for their growth.

There is now general consensus that the major consumers of suspended bacteria are small (less than 5 μm) nonpigmented (as well as some pigmented) flagellates (Haas & Webb, 1979; Fenchel, 1982; Bird & Kalff, 1986; McManus & Fuhrman, 1986; Sanders & Porter, 1988; Wikner & Hagstrøm, 1988; Sanders et al., 1992), and to a lesser extent ciliates (Børsheim, 1984; Sherr & Sherr, 1987) and dinoflagellates (Lessard & Swift, 1985). Evidence also identifies viruses as major controlling factors of bacterial populations (Proctor & Fuhrman, 1990). In sediments where bacterial concentrations are much higher than they are in the water column, ciliates and invertebrates, such as meiofaunal metazoans and polychaetes, routinely ingest large amounts of bacterial biomass (Fenchel, 1967, 1968; Capriulo, 1990).

Fate of primary production

Although micro-algal composition varies over time between net *versus* nano-sized species in many marine waters, investigations in both temperate and tropical, neritic and oceanic, waters have demonstrated that nanoplankton (and smaller) forms are often responsible for 80–99% of the observed phytoplankton productivity (Ryther, 1959; Anderson, 1965; Yentsch & Malone, 1971). Additional evidence suggests that a major portion of the chlorophyll in many marine/estuarine waters is bacterial in size (e.g. less than or near 1 μm) (Johnson & Sieburth, 1979, 1982; Morris & Glover, 1981). This production is utilized primarily by zooplankton (macro and micro forms) with microalgal size, quality and species composition determining which zooplanktonic forms predominate (Capriulo, 1990).

Tiny copepod crustaceans are the most abundant multicellular animals in the ocean water column. More than 80% of the mesozooplankton in LIS during most of the year are copepods (Deevey, 1956; Anonymous, 1976–83). As free swimming suspension feeders, they influence phytoplankton and protozoan populations (Pechnick, 1985; Lutz, 1986; Walker & Peterson, 1991). They are preyed upon in turn by larger organisms including ctenophores and fish (Deevey, 1956; Johnson, 1987; Kiørboe et al., 1988; Peterson et al., 1992; Verheye et al., 1992; Nielsen et al., 1993). Adult herring, anchovies, menhaden, and juveniles of other species that enter LIS are well adapted for feeding on copepods (Deevey, 1956). Sloppy feeding and egestion by copepods supply dissolved organic matter (DOM) and small fragments to the microbial pathway (Capriulo et al., 1988; Peduzzi, 1992), and the inorganic nitrogen copepods excrete supports primary production (Glibert et al., 1992). In addition, copepods contribute to the vertical flux of carbon by egesting rapidly sinking fecal pellets (Butler & Dam, 1994), and (or) by performing diel vertical migrations (Longhurst et al., 1990).

The broad range of water temperatures (0–25 °C) in temperate LIS induces seasonal dominance of distinct warm-water (summer–fall) and boreal (winter–spring) copepod communities, both containing species adapted to the reduced and variable salinity of estuarine waters (Deevey, 1956; Peterson, 1986). Most copepods found in LIS are also present in nearby neritic waters, but there are species present to the east in Block Island Sound that appear to be excluded by the reduced salinity of LIS. Thus the numerically important species in LIS are few: *Acartia tonsa* (summer–fall), *Acartia hudsonica* (winter–spring), *Temora longicornis* (winter–spring), *Pseudocalanus minutus* (winter–spring), *Paracalunus crassirostris* (summer–fall), *Centropages* sp, and *Oithona* sp. (summer–fall) (Deevey, 1956).

Predation upon copepods

Predation can temporarily decimate copepod populations (Johnson, 1987; Peterson et al., 1992; Verheye et al., 1992). Ctenophores can be voracious seasonal predators in LIS between the months of June and September. Copepod populations in the summer tend to vary inversely with ctenophore populations. Abundant ctenophores preyed upon the nauplii of *Paracalanus crassirostris* at a rate of approximately 25% d^{-1} in September 1985; however, in 1986, a

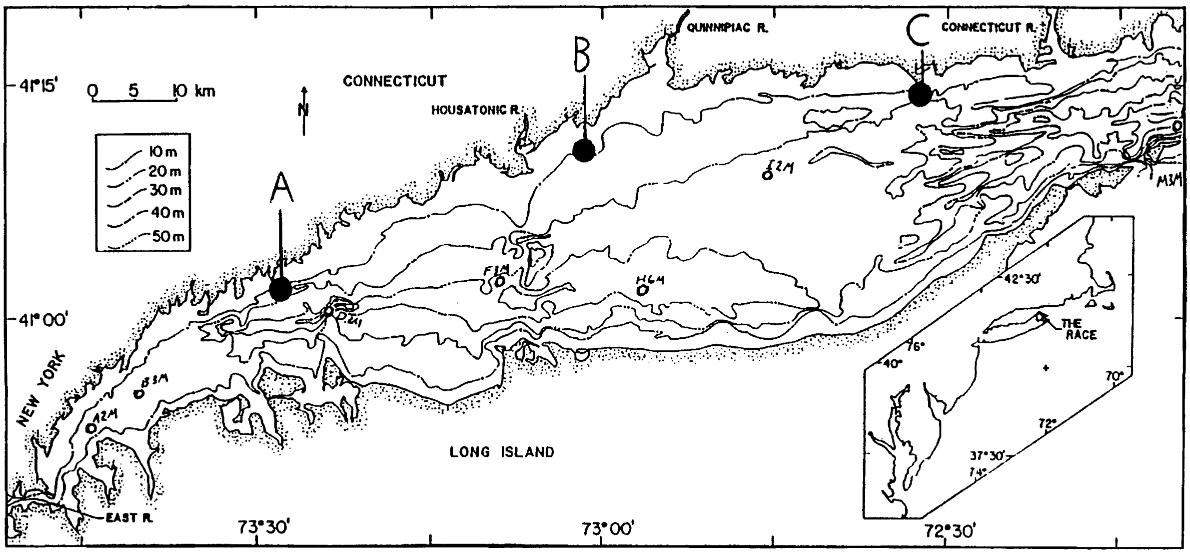

Figure 1. Map of the Long Island Sound (U.S.A.) estuary, showing the 3 study stations: A = Stamford, CT, B = Milford, CT and C = Hammonasset, CT.

year of fewer ctenophores, the nauplii of *Paracalanus crassirostris* were preyed upon by abundant *Acartia tonsa* instead (Johnson, 1987). The large predatory copepod, *Labidocera aestiva*, is never a major component of the zooplankton in LIS (Deevey, 1956; Johnson, 1987). There has been conjecture about fish limiting copepods in LIS (Peterson, 1985), and the Chesapeake Bay (Purcell et al., 1994). Anchovies, herring, and menhaden that are known to consume copepods can be quite abundant in LIS (Deevey, 1956).

Long Island Sound

Long Island Sound (LIS) is a temperate estuary with an area of about 900 nautical square miles, bordered by the states of Connecticut and New York (Fig. 1). In LIS the salinity ranges between 23 and 31 practical salinity units (PSU), the temperature ranges between 0 and 25 ° C, and the average depth is 20 m (Riley, 1955).

The most comprehensive study of the biological oceanography of Long Island Sound was carried out by a group of scientists working with Gordon Riley in the 1950s (Riley et al., 1956, 1959). This group analyzed the physical and chemical oceanography of Long Island Sound as well as the population/ecological dynamics within and among the phytoplankton, zooplankton and pelagic fish eggs and larvae. Nutrient dynamics, the role of organic matter and the biology of bottom communities were also considered. This work remains as vital to our understanding of the Long Island Sound estuary today as it was when first reported.

The Riley group estimated the annual primary fixation of carbon Sound-wide to be about 470 g cm^{-2} yr^{-1}. Of this number more than half is used in phytoplankton respiration with 205 g cm^{-2} yr^{-1} remaining available for other uses. Riley et al. further estimated that 26% of the available production goes to macrozooplankton, 31% to benthic fauna and flora and by mass balance subtraction concluded that 43% goes to microzooplankton and bacteria. The above work also reported data on phosphate, nitrate, and chlorophyll concentrations, zooplankton volume and zooplankton, phytoplankton and larval fish species composition and abundance.

Although a number of discrete studies on various aspects of Long Island Sound's biological community have been carried out over the years since Riley et al.'s work (1956, 1959), few of these have been published in the refereed scientific literature. The NOAA, National Marine Fisheries Service has conducted bacterial and phytoplankton research as they relate to shellfish ecology (Tettelbach et al., 1984; Brown et al., 1988; Wikfors in prep., Wikfors et al., in prep). These studies were generally of short duration and limited geographic range but do show clear seasonal and geographic differences in abundance and/or taxonomic composition of both bacterial and phytoplankton communities in Long Island Sound.

In the late 1970s and early 1980s Capriulo & Carpenter (1980, 1983) conducted research to examine the details of phytoplankton abundance and community structure, and tintinnid ciliate grazing on primary production in central Long Island Sound. That work found that the microzooplankton removed up to 41% of the standing crop per day, and the tintinnid ciliates 27% of the annual primary production. Also, tintinnid ciliates exhibited community ingestion rates equal in order of magnitude to the copepod community. With respect to phytoplankton, small nanoplankton often dominated microalgal communities which likely explains the success of the ciliates. The 1983 study only examined tintinnid feeding impact, yet naked oligotrichous planktonic ciliates often outnumber the loricate forms (Smetacek, 1981; Montagnes et al., 1988) in nature, and by as much as 5× in Long Island Sound (McManus, unpublished data and pers. comm; Capriulo & Pellet, unpublished data). This suggests that the actual feeding impact of ciliates is likely to be much higher than currently estimated and therefore probably is greater than that of the copepod community.

Welsh & Eller (1991) examined, physical, chemical and biological factors controlling oxygen depletion in western Long Island Sound. In the mid-1980s Peterson (1985), Peterson & Bellantoni (1987), Monteleone et al. (1987), and Peterson (unpublished data) conducted a seasonal study of the phytoplankton, copepods, larval fish and nutrient dynamics of central Long Island Sound patterned after Riley's et al. 1950s work (1956, 1959). This work ignored, as did Riley's et al. the bacterial, heterotrophic flagellate and other protozoan components of the Long Island Sound ecosystem. In fact, no comprehensive study of the microbial loop components of the Long Island Sound ecosystem has ever been carried out.

Annual plankton cycle in Long Island Sound

Distinct annual cycles in concentrations of nitrogen, chlorophyll, and copepods occur as a consequence of seasonally changing light intensity, day length, and temperature in temperate Long Island Sound. The algae are light limited in the winter (Riley, 1969). Inorganic nitrogen concentrations are high due to regeneration at the end of the previous season (Harris & Riley, 1956; Harris, 1959). As light increases, diatom blooms may be triggered by any slight increase in the stability of the water column due to the combined influences of temperature and salinity on density. Winter blooms begin as early as January, mostly in February, but can be delayed until March or later (Riley, 1969; Capriulo et al., this study). During the bloom, inorganic nitrogen decreases and particulate nitrogen increases (Harris & Riley, 1956; Harris, 1959). The bloom ends when nutrients are gone, when self-shading becomes too intense, when there is adverse weather, or when grazing becomes too intense (Riley, 1969). Copepods are generally not very abundant in winter (Deevey, 1956). A substantial proportion of the bloom settles to the bottom and decomposes, releasing inorganic nitrogen back into the water column (1956; Harris 1959; Riley & Conover, 1967; Harris & Riley, Riley, 1969).

In the spring, there are moderate pulses in phytoplankton abundance, and blooms of either diatoms or flagellates may happen in May or June if enough nitrogen is available (Riley & Conover, 1967). Flagellates begin to replace diatoms as the season progresses. Flagellates are favored over diatoms by stratification and low inorganic nitrogen (Riley & Conover, 1967; Demers et al., 1978; Margalef, 1978; Mann, 1982; Peterson, 1986; Mann & Lazier, 1991; Kiørboe, 1993). A thermocline forms at 6–8 m, usually by sometime in June (Welsh & Eller, 1991). The concentration of inorganic nitrogen becomes nearly undetectable as spring progresses, and rapid recycling of inorganic nitrogen becomes the keystone of primary production (Harris & Riley, 1956; Harris, 1959; Riley, 1969; Peterson, 1986). The winter-spring copepod species increase to their peak abundance through April and May (Deevey, 1956).

During the summer, phytoplankton populations are nutrient limited until late in the season, and remain small compared with bloom conditions; however, primary production may actually be quite high (Riley, 1956b). Dinoflagellates have largely replaced diatoms (Riley & Conover, 1956). The winter–spring copepods are replaced by summer–fall varieties, which peak in abundance around August, if not decimated by predators such as ctenophores (Deevey, 1956; Conover, 1956; Johnson, 1987). Decreasing solar radiation and water column stability late in the summer force reductions in primary production such that a surplus of nitrogen supply over utilization occurs again (Harris & Riley, 1956; Harris, 1959).

In the fall, or even as early as late August, diatom blooms can be triggered when the fall destabilization of the water column is temporarily interrupted by some favorable combination of temperature, salinity, and light winds (Riley, 1959). Fall blooms tend to vary a lot from year-to-year, but they occasionally interrupt the autumnal increase in inorganic nitrogen concentrations (Riley, 1959). Phytoplankton remain limited by light, and dissolved inorganic nitrogen concentration peaks in November or December (Harris, 1959). The summer–fall copepods give way to winter–spring varieties in November and December (Deevey, 1956).

Estuarine copepods have been shown to feed very inefficiently on small phytoplanktonic forms when compared to planktonic ciliates offered identical food (Capriulo & Ninivaggi, 1982). Copepods exhibit higher ingestion rates when fed large *versus* smaller algae (e.g. 30 μm *versus* 10 μm nodal sizes) (Mullin, 1963; Frost, 1972; O'Connors et al., 1976, 1980). O'Connors et al., (1980) found for the Long Island Sound copepod *Temora longicornis* feeding on natural food, that maximum ingestion rates increased linearly by a factor of 3.5× as food size increased from 5 to 30 μm (i.e. flagellate to diatom dominated). Capriulo & Ninivaggi (1982) noted that the copepod/tintinnid ingestion rate ratio varied from 147:1 to 24:1 as the modal natural food size shifted from 15 μm to 4 μm, while the maximum filtration rate remained constant at about 75:1 under both conditions. These results indicate that protozooplanktonic ciliates become increasingly more important as food sizes shift towards smaller sizes. A major consequence of such a shift towards smaller algae and protozoans is a lengthening, by one or more steps, of the food chain leading from primary producers to fish. Protozoan gross growth efficiencies vary by species from a low of about 2% to a high of 82% (Caron & Goldman, 1990). These numbers suggest an average protozoan ecological transfer efficiency of approximately 30%. Thus, for each step added to the planktonic food chain by enhancement of the protozoan components of the microbial loop, a 70% reduction of resultant finfish/shellfish production can be expected. Changes in the zooplankton composition resulting from such trophodynamic shifts are important to finfish and shellfish production.

Copepods appear to feed most effectively on microplankton (plankton >10 μm) (Frost, 1972; O'Connors, 1980; Capriulo & Ninivaggi, 1982; Dam, 1989). Production within nanoplankton and picoplankton (plankton < 10 μm) may be more likely to enter the microbial pathway (Capriulo & Ninivaggi, 1982). Moreover, differences in the abundance of copepods in a food limited environment might correspond with the concentration of microplankton. The literature suggests that copepods are food limited in LIS, perhaps not all the time, and maybe not everywhere, but at least intermittently (Conover, 1956; O'Connors et al., 1980; Peterson, 1986; Dam, 1989). Copepod weight, length, abundance, species distribution, and rate processes such as growth, ingestion, excretion, and respiration vary with food availability (Frost, 1972; Conover & Huntley, 1980; Huntley & Boyd, 1984; Omori & Ikeda, 1984; Ikeda, 1985; Kiørboe et al., 1985; Berggreen et al., 1988; Cowles et al., 1988; Kiørboe, 1989; Dam & Peterson, 1991; Bautista et al., 1992; Bautista & Harris, 1992), temperature (Conover, 1956; Dam, 1989), and salinity (Deevey, 1956).

Potential altered food web structure in Long Island Sound

Several studies have shown that changes in water column chemistry (e.g. dissolved oxygen content, nutrient concentrations and relative ratios such as N/P or N/Si, presence of toxicants such as heavy meals, PCB's, DDT's etc.) alter the species composition of phytoplankton, bacteria, protozoa as well as metazoans, in nature (Thomas & Seibert, 1977; O'Connors et al., 1978; Sanders et al., 1987; Fenchel et al., 1990; Søndergaard et al., 1991). For phytoplankton, taxonomic composition, as well as gross productivity are dependent on absolute amounts and ratios of various inorganic nutrients, especially nitrogen, phosphorus, and silicate. Nitrogen and phosphorus are required by all phytoplankton, and absolute quantities of these two nutrients are generally thought to limit gross production of microalgae in marine and freshwater ecosystems (Harris, 1986). For a combination of reasons related to Molybdenum availability as well as N/P ratios, nitrogen tends to be the limiting nutrient in marine ecosystems (Howarth & Cole, 1985). Therefore, anthropogenic inputs of nitrogen stimulate algal growth in systems such as Long Island Sound. Decreasing the N input to Long Island Sound would thus be expected to reduce primary production of phytoplankton biomass. The ratio of N/P may vary from west to east because of higher sewage-derived nutrient loadings in the west. This ratio may be important in controlling the species composition of phytoplankton communities (Sakshaug et al., 1983; Søndergaard et al., 1991).

Although the absolute concentration of N is generally implicated in the limitation of gross production in

Long Island Sound, the ratio of N/Si may be the most important factor regulating taxonomic composition. Diatoms require Si for incorporation into their cell walls, to grow (Harris, 1986). The dominance of diatoms in the spring 'bloom' in Long Island Sound is well documented, as is the succession later in the season to other algal taxa, especially flagellates (Conover, 1956; Capriulo & Carpenter, 1983; Wikfors, in prep.). Research into the question of why diatoms dominate the spring phytoplankton has revealed that small diatoms possess higher nutrient uptake and growth rates than larger diatoms and other algal taxa (Geider et al., 1986; Sanders et al., 1987; Hulburt, 1988; Furnas, 1990). Replacement of diatoms by other typically small-sized algal species follows removal of the available silicate from solution by the diatoms and subsequent sinking of Si-containing cells out of the euphotic zone (Harrison et at., 1986; Anderson & Nival, 1989). Without Si, diatoms cannot sustain rapid growth rates and other phytoplankton taxa with slower growth rates become dominant, until breakdown in stratification remixes Si into the euphotic layer. A higher N/Si ratio (due to eutrophication) in western Long Island Sound should result in a shorter period of diatom dominance relative to the eastern Sound, and a longer season of flagellate dominated production in the west (Wikfors, in prep.). For reasons already discussed, this should result in reductions in regional finfish and shellfish populations and productivity.

Additionally, the 'excess' primary production stimulated by N eutrophication results in more release of algal organic exudates (Hagstrøm et al., 1979; Fuhrman & Azam, 1982) which stimulate bacterial production and therefore production of protists and bacterial grazers. Heightened bacteria and bacteria-grazer production results in lower dissolved oxygen concentrations due to high community respiration rates. Also, if the added algal biomass goes underutilized by grazers, then additional bacterial growth is stimulated via the decomposition of dead algal biomass both in the water column and the benthos. Such a result would strip more oxygen from the water column and benthos resulting in hypoxia or anoxia (Welsh & Eller, 1991). Such hypoxia/anoxia itself results in species compositional changes (Fenchel et al., 1990) superimposed on those already realized, thus further stimulating the microbial loop and driving down microcrustacean, finfish, and shellfish production.

The Connecticut Department of Environmental Protection has identified a west–east eutrophication gradient in LIS (unpublished and internal agency data). One would expect copepod stocks to respond to these gradients if eutrophication altered the amount of food available to them within the range of food limitation. Riley (1955) suggested an east-to-west increase in zooplankton in Long Island Sound; however, in a later paper, Riley (1959) implied that the data he had available did not indicate a clearly significant increase. Changes in copepod abundance, biomass, individual weight, and species have been noted to occur over gradients of eutrophication in other parts of the world (Painting et al., 1993; Uye, 1994). The switch to secondary treatment of the sewage entering LIS has shifted the form of the added nitrogen from organic forms to inorganic forms over recent decades, which may have affected the food web.

Whether nitrogen addition yields increased microphytoplankton, or increased nanophytoplankton (PNANS), is mediated by mixing, stratification, and light, as well as by the form of the nitrogen addition (Riley & Conover, 1967; Margalef, 1978; Mann, 1982; Demers et al., 1986; Peterson, 1986; Legendre & Le Fèvre, 1989; Mann & Lazier, 1991; Kiørboe, 1993). The microphytoplankton in LIS tend to be diatoms, and diatom populations are generally disfavored by stratification because they need a certain amount of turbulence to keep them mixed up into the euphotic zone. PNANS, on the other hand, are smaller and so they sink more slowly, and they may employ flagella for depth control (Demers et al., 1986; Mann & Lazier 1991). In addition, when inorganic nitrogen concentrations are high there tend to be more diatoms, and when inorganic nitrogen concentrations are low there tend to be more PNANS (Kiørboe, 1993). PNANS acquire nitrogen more efficiently than diatoms because they have a higher surface area to volume ratio, because their mobility may allow them to take better advantage of heterogenous nutrient concentrations, and because they are more likely to utilize organic nitrogen to satisfy part of their nitrogen needs (Conover, 1956; Valiela, 1984; Parsons et al., 1984; Mann & Lazier, 1991). Logically, eutrophication in LIS should favor PNANS and diatoms both over the course of the year, since conditions range from a mixed water column with high inorganic nitrogen, to a stratified water column with low inorganic nitrogen. Other factors to consider are that the euphotic zone may narrow with eutrophication due to shade produced by the additional cells (Beeton, 1969), and that a potential exists for silicon abundance to limit diatoms, but not PNANS, since diatoms require silicon to form frustules, and PNANS do not.

Changes in copepod populations tend be separated in time from changes in phytoplankton populations (Legendre & Le Fèvre, 1989; Walker & Peterson, 1991). This is because a month or more may pass before copepod populations recruit new members, whereas phytoplankton and heterotrophic microbes can double their number in hours or days (Conover, 1956; Atlas & Bartha, 1987). The implication here is that the copepod and microbial pathways partition food based not only on cell size, but also based on the match or mismatch between the temporal characteristics of food availability and those of copepod population dynamics.

Nutrient overloading has occurred in the western Long Island Sound, and hypoxic conditions in summer have been routinely encountered, particularly in the bottom waters of deeper basin sites (Long Island Sound Study Annual Report 1988, 1989/1990, and 1990) due to higher algal biomass, with maxima in the 30 μg Chl *a* l^{-1} or more range in the western Sound, as compared to 10 μg Chl *a* l^{-1} in the east (Olsen, 1975; Cosper, SUNY Stony Brook, Marine Science Research Center, unpublished data).

The fundamental question which arises from this is why the excess primary production results in hypoxia via decomposition pathways rather than enhanced secondary production of a quality leading to more fish and shellfish production. We hypothesize that the answer to this question lies in the fact that the central to western Long Island Sound food web dynamics have been shifted more towards a microbial loop dominated system as compared with the more 'traditional' food web dynamics of the central to eastern Long Island Sound. If such a fundamental shift has occurred, then both finfish and shellfish populations might be negatively affected.

Material and methods

To address our hypothesis and the major research questions of this work, the following parameters were measured at monthly intervals at three 10 meter, near shore stations (Fig. 1), on the Connecticut, U.S.A. side of Long Island Sound.

1. nutrient concentrations (ammonia, nitrate, nitrite, dissolved organic nitrogen (DON), particulate organic nitrogen (PON), phosphate and biologically active silicate), temperature, salinity and density, vertical CTD profiles and vertical oxygen profiles)
2. bacterial densities
3. bacterial growth rates (estimated as FDC)
4. total and size-fractionated chlorophyll *a* and phaeopigment concentrations (total, <10 μm, 10–20 μm, >20 μm)
5. phytoplankton concentrations and species composition
6. heterotrophic nanoplankton (HNAN) densities
7. photosynthetic nanoplankton (PNAN) densities
8. protozooplankton (chiefly ciliates) species composition
9. macrozooplankton concentrations and species composition (excluding ctenophores)
10. first approximation larval fish concentrations and species composition.

Schedule of sampling

The three stations were spaced at intervals of about 45 k along the 10 m isobath of the Connecticut shoreline, adjacent to the towns of Stamford and Milford, and Hammonasset State Park (Fig. 1). Sampling was performed monthly (occasionally weekly), typically between 0830 h and 1200 h on two consecutive days, two stations on one day and one on the next. Sampling took place for 3 years (June 1992–May 1995) at Stamford and Milford, and 2 years (June 1993 – May 1995) at Hammonasset. With an occassional rare exception 3 research vessels were used for this work: the R.V. Oceanic of the Maritime Aquarium at Norwalk, for Station A, the R.V. Shang Wheeler of NMFS, Milford for Station B, and the R.V. Libinia, UCONN Marine Science Institute for Station C. These stations are representative of the near shore western, central and eastern Long Island Sound regions, respectively.

Water sampling

Approximately 15 l of water were taken at each station from each of two depths. A model-1080 General Oceanics Go-Flow, teflon lined water sampler was used to take water from 1 m off the bottom (in a 10 m depth water column). Surface water was collected using a plastic bucket on a rope, except that water for Winkler dissolved oxygen titration was taken using the Go-Flow sampler to avoid exposure to air. Two or three casts per depth were required to obtain sufficient quantity of sample. They were combined as one sample in large Nalgene carboys. Water was dispensed from the carboy for particulate filtration, and to various bottles for a multitude of analyses. All containers, including the carboys and buckets, were prepared by rinsing with 10% hydrochloric acid, distilled deionized water, and sample.

Physical parameters

Temperature, salinity and oxygen were measured using a combination of instruments including: a mercury thermometer, an American Optical refractometer, an Applied Microsystems STD-12 profiling unit which logged depth, temperature, salinity, and sigma-t to a computer, a YSI Model 33 SCT meter, a YSI Model 58 oxygen meter and Winkler titration (Strickland & Parsons, 1972).

Nutrient analyses

Subsamples for all nutrient analyses were transferred to acid washed, DDW rinsed BOD bottles and were placed on ice prior to same day analyses. Samples were transported from the field to the lab in an ice-filled cooler.

Nitrate, nitrite, ammonia, & phosphate determinations

Surface and bottom water nitrate, nitrite, ammonia and phosphate concentrations were determined using established, standard methods (Parsons et al., 1984). All determinations were carried out immediately after each research cruise, upon return to the laboratory. Silicate, DON and PON analyses were performed at a later date on samples that had been kept frozen (Parsons et al., 1984). Absorbences for all 3 species of dissolved inorganic nitrogen were measured using a research grade, Carey 118, spectrophotometer. Concentrations are presented as μg-at N per liter.

TDN and DON determinations

Dissolved organic carbon (DON) was measured after the method of Solorzano and Sharp as outlined in Parsons et al. (1984). Determinations were carried out in triplicate on 100-ml seawater sub-samples drawn from the original water samples, and tranferred to acid washed and distilled deionized water rinsed BOD bottles which were kept in ice filled coolers for transport back to the laboratory. All samples were pre-filtered through 0.45 μm millipore filter and kept frozen until analyses were carried out.

Total dissolved nitrogen analyses were carried out on samples collected between August 1993 and May 1995. The operational definition of TDN was that it pass a Millipore 0.45-μm membrane filter. Equipment and glass-ware contacting TDN samples were triple-rinsed with 10% hydrochloric acid prior to use. Samples for TDN were kept on ice until filtered, and then frozen in 250-ml glass Wheaton-bottles within several hours of when they were taken. Analysis was typically within a week, and performed according to method 2.1 of Parsons et al. (1984). An alkaline persulfate digestion in an autoclave oxidized all forms of nitrogen to nitrate (NO_3). The resulting NO_3 was measured according to method 1.1 from Parsons et al. (1984) by reduction to nitrite (NO_2) in a cadmium column, and the addition of color forming reagents prior to photometric determination with a Hewlett Packard spectrophotometer.

Inorganic nitrogen was also analyzed. Method 1.1 of Parsons et al. (1984) was used to determine NO_x, method 1.3 of Parsons et al. (1984) was used to determine NH_3. Ammonium was oxidized to nitrite with hypochlorite in alkali using a large excess of potassium bromide as a catalyst, and measured photometrically as in method 1.1. Dissolved inorganic nitrogen (DIN) was calculated by summing NO_x and NH_3. Dissolved organic nitrogen (DON) was calculated as the difference between TDN and DIN.

Particulate organic nitrogen (PON) determinations

Particulate organic nitrogen (PON) was measured in triplicate 100-ml seawater samples pre-filtered with 200 mesh Nitex monofilament screen. Particles were collected on 25-mm glass fiber filters with nominal retentions of 1 μm (Whatman GF/B), making the range of particle size included in PON measurements 1–200 μm. Seawater samples were transported to the laboratory on ice and filtered. Filters were stored at −80°C, wrapped in aluminum foil, until analyzed. The analytical method used for PON determinations was a heated biuret-Folin protein assay (Dorsey et al., 1977, 1978), modified according to Wikfors et al. (1984), read with a Beckman DU-40 Spectrophotometer. The use of a protein–nitrogen assay to represent total particulate N was justified based upon the assumption that non-protein PON would be very small compared with protein PON. It should be noted that PON values represent both photosynthetic and nonphotosynthetic organisms, and non-living particles within the size range selected.

Particulate nitrogen was analyzed beginning with samples from the June 1993 cruise through May 1995. PON was measured as that retained on Whatman GF/F glass-fiber filters which capture particles nominally >0.7-μm. Filtration was typically completed within 1–1.5 h of water sampling, and filters kept on ice, until frozen at −70 °C later that day. The procedure was that of Wickfors et al. (1984) modification of Dorsey et al. (1977, 1978). Nitrogen was extracted in reagents at 100 °C for 100 min, and measured on a spectrophotometer.

Total nitrogen determinations

Total nitrogen (TN) was calculated by summing TDN and PON.

Biologically active silicate determinations

Dissolved, biologically-available silica (BSi) was analyzed in seawater samples using the molybdate colorimetric method of Strickland & Parsons (1977). The published method reports precision of ±0.14 at 10 μM; However, in our laboratory, the method was found to be linear to 0.01 μM, with accuracy of ±0.002 at 0.1 (Wikfors, unpublished data). The method was found to be non-linear, however, above 60 μM. Hence, samples containing 40+ μM BSi were diluted with deionized water and re-analyzed. Seawater samples were transported from the field on ice and frozen at −20°C until analysis within one week of collection (experiments found no significant differences in BSi values for samples analyzed immediately and those stored for up to 2 weeks at −20 °C; Wikfors, inpubl. data). Frozen samples were thawed and centrifuged at 2800 ***g*** for 20 min. prior to analysis. Extinctions were read with a Beckman DU-40 Spectrophotometer.

Phytoplankton identifications & semi-quantitative population estimates

Seawater samples for live observation were transported to the laboratory on ice. On the same or next day (usually within 2 h of collection, always within 12 h), the live samples were concentrated (about 200×) by centrifuging gently (10 min. at 1000 G) and evaluated for living phytoplankton using epi-fluorescence microscopy at 400 and 1000 magnifications. The value of observing live samples in this semi-quantitative way is that living cells could be differentiated from empty cell walls by chlorophyll *a* fluorescence in living cells. Further, photosynthetic cells less than 2 μm in size, which are virtually impossible to visualize or quantify in fixed samples, could be estimated. Percentage estimates of dominant taxa (to the lowest taxonic level possible) were made based upon the contribution of each taxon to the fluorescent particles in a few representative fields.

Phytoplankton counts

For 1994, counts were made of phytoplankton in Lugol's iodine-fixed samples from surface waters of Stamford and Hammonasset. Two liter samples were concentrated by gravity settling, through several steps, to 10–50 ml, depending upon cell density. Four subsamples of each concentrated sample were counted in an Improved Neubauer Hemocytometer (American Optical, Bright Line). Live-cell records were consulted to determine if fixed cells were likely to have been photosynthetically-active at time of collection. Counts were converted to units of cells per liter.

Chlorophyll a analyses

All water collected for chlorophyll analyses was left unscreened or was prescreened through either 10 μm or 20 μm nitex mesh and then filtered through Gelman Type A/E glass fiber filters (25 cm diameter, pore size ca. 0.5 μm). The filtration manifold was connected to a series of 2 side arm flasks, the second of which was connected to a vacuum pump set typically at 4 psi (and never above 10 psi). The apparatus was assembled for filtration of triplicate 15 ml samples each of unscreened, 10 μm mesh screened or 20 μm mesh screened water. Appropriate subtractions allowed determination of total greater than 20 μm sized, 10–20 μm sized and less than 10 μm sized chlorophyll *a* concentrations. Following filtration, filters were removed with forcepts (a necessary technique to insure that chlorophyll is not acidified via contact with hands) folded in half, wrapped in aluminum foil and placed on ice for transport to the laboratory. One and a half to 2 h after collection, samples were either fluorometrically analyzed or frozen for analysis within a few days to 2 weeks later. Each filter was removed with forcepts and placed in borosilicate glass test tubes (which were prepared with 90% grade A-19 Fisher acetone diluted with distilled deionized water) and ground up in 90% acetone using a teflon grinder attached to a variable speed drill. Glass fiber residue was removed via filtration through type A/E glass fiber filters prior to fluorometric determinations of chlorophyll using a Turner 111 fluorometer for June 1992–May 1993 samples and a Turner Designs 10-AU fluorometer for the June 1993–May 1995 samples. Chlorophyll extract acetone solutions were diluted once more to 10 ml with 90% acetone and transferred to a cuvette. Once fluorometric measurements were recorded 2 drops of a 0.5% HCl solution were added to allow for phaeopigment determinations. This method represents a slight modification (i.e. grinding of filters for rapid chlorophyll extraction) of standard fluorometric methods (Parsons et al. method 4.3, 1984; Mantoura et al., 1990; Bidigare, 1991).

Bacterial, HNAN & PNAN analyses

Water samples for microbial analyses were collected as for nutrient, chlorophyll, phytoplankton and protozooplankton determinations. Subsamples were then transferred to a primed, graduated cylinder and 10 ml of sample were transferred to a sterile screw cap vial containing 0.5 ml of 37% formaldehyde. Two replicate vials from each depth were created and transported on ice back to the laboratory for subsequent analyses. Samples were kept at 4 degrees centigrade in the dark and typically analyzed within days to 2 weeks. An occassional sample was analyzed 4 weeks after collection.

Bacterial abundances were determined using epifluorescence microscopy and the acridine orange staining method of Hobbie et al. as described in Parsons et al. (1984). Nuclepore (0.22 μm pore size and 25 cm diameter) filters were used. Bacterial production was estimated using the FDC technique of Hagstrøm (1979) on acridine orange stained cells. Heterotrophic (HNAN) and photosynthetic (PNAN) densities were also estimated using a single filter stain technique (i.e. following the Hobbie et al. procedure for bacterial counts). The preferred method for the HNAN and PNAN determinations is a double filter (one AO stained and the other unstained) technique (Sherr et al., 1994). Subtraction of autofluorescing PNAN's on the unstained filter from the total counts on the stained filter allows for estimation of the HNAN density by difference. In a single AO stained filter technique, some HNAN's weakly fluoresce in the red range due to RNA interaction with the AO stain, giving some level of false positives for PNAN's and an underestimation of HNAN's. Due to the large quantity of parameters (most of which have time constraints related to analyses) measured on each series of sampling cruises, we could not routinely employ this double filter method. The replicate single filter AO stained samples were therefore used for approximate determinations of HNAN and PNAN densities. To determine the magnitude of potential errors resulting from this technique, we carried out the double filter procedure on a number of occassions to determine the difference in estimates gained from the 2 methods. From this a gross correction term was calculated (Table 1).

Analyses were carried out with an Olympus BH-2 Phase Contrast Epifluorescence Microscope fitted with a 100 W mercury vapor lamp, under a 100× oil plan-achromat Olympus objective. The microscope was also fitted with a whipple disk in one of the oculars, to facilitate in enumerating the bacterial cells.

While counting bacteria, those cells which were observed to be in the process of division (evident as a clear invagination of the cell wall without a distinct separatory space between cells) were noted and recorded as a percentage of the total count.

A total of four random fields were counted, and the total number of cells in each field were recorded. An additional 20 random fields were then observed, and the total number of heterotrophic (HNANs) and phototrophic (PNANs) nanoplankton were recorded. HNANs, like bacteria, because they are nonphotosynthetic and contain no chlorophyll, fluoresced bright green. HNANs were distinguished from bacterial cells by their size (roughly 2–5 times the size of bacterial cells). PNANs although typically the same size as HNANs, were clearly distinguished from HNANs and bacterial cells by the presence of chlorophyll which fluoresced bright red-orange. Flagella were sometimes clearly discernable, whereas at other times they were absent. Each cell count was then averaged together and abundance calculated. Two replicate counts were made per vial (2 vials were collected for each depth) for a total of four counts per depth and eight counts per station. Once the slides were counted, and the results recorded, the slides were placed in a microscope slide holder, labelled and placed in the freezer for reference at a later date if needed.

Protozooplankton analyses

Two liters of water from each Niskin bottle collection were fixed in Lugol's solution in a large graduated cylinder and allowed to settle by gravity for 1 or 2 weeks. Supernatant was aspirated off and samples concentrated to 100 ml. One liter from the Niskin bottle sample was fixed in Bouin's fixative (a saturated solution of picric acid in formaldehyde, with the addition of 1% final concentration glacial acetic acid), placed in a graduated cylinder and also concentrated

Table 1. Comparison of single and double filter fluorescence microscopy methods for estimating photosynthetic nanoplankton (PNAN) and heterotrophic nanoplankton (HNAN) densities, showing over and under-estimate errors and associated correction factors with respective standard deviations

PHAN density (× 10^4)			HNAN density (× 10^3)		
Single	Double	Corr. ×	Single	Double	Corr. ×
4.67	5.22	1.12	1.27	3.12	2.5
2.88	2.91	1.01	1.42	3.12	2.2
1.52	1.06	0.70	0.28	4.83	17.2
1.43	0.79	0.55	0.43	6.80	15.8
2.37	1.52	0.64	1.42	9.91	7.0
2.20	1.66	0.75	0.85	6.23	7.3
1.72	0.98	0.57	0.90	4.84	5.4
1.04	1.06	1.02	0.43	1.84	4.3
1.55	0.98	0.63	0.85	6.52	7.7
1.12	0.84	0.75	0.85	3.69	4.3
0.73	0.76	1.04	0.71	1.42	2.0
0.58	0.63	1.08	1.13	1.23	1.1
0.31	0.20	0.64	0.28	2.13	7.6
0.57	0.27	0.47	0.99	4.53	4.6
0.91	0.44	0.48	0.85	5.52	6.5
0.94	0.53	0.56	0.57	4.68	8.2
0.92	0.43	0.47	0.71	6.51	9.2
0.38	0.36	0.94	0.57	1.14	2.0
1.28	0.96	0.75	0.85	4.40	5.2
0.92	0.50	0.54	1.14	5.39	4.7
5.12	3.73	0.73	1.13	15.01	13.3
2.82	2.40	0.85	1.42	7.53	5.3
1.25	0.95	0.76	0.28	3.26	11.6
1.62	0.98	0.60	0.43	6.81	15.8
2.71	1.90	0.70	0.42	8.50	20.2
2.06	2.13	1.03	0.57	1.99	3.5
1.56	1.02	0.65	1.70	6.24	3.7
1.25	0.75	0.60	0.85	5.67	6.7
1.16	0.91	0.78	0.71	3.40	4.8
1.14	0.92	0.81	0.99	3.12	3.2
0.75	0.52	0.69	0.85	2.41	2.8
0.53	0.48	0.91	0.61	1.42	2.3
0.44	0.17	0.39	0.85	3.53	4.2
0.43	0.30	0.70	0.57	1.55	2.7
0.65	0.19	0.29	0.43	5.10	11.9
0.64	0.16	0.25	0.57	5.40	9.5
0.99	0.61	0.62	0.14	4.95	35.3
0.77	0.40	0.52	0.57	3.97	7.0
0.78	0.51	0.65	1.42	4.10	2.9
0.65	0.58	0.89	0.71	1.56	2.2
2.20	1.42	0.64	0.99	8.92	9.0
1.82	1.96	1.08	0.85	2.13	2.5
0.78	0.67	0.86	0.28	2.13	7.6
0.89	0.84	0.94	0.14	1.98	14.1
2.25	1.73	0.77	0.14	6.09	43.5
2.61	1.63	0.62	0.85	10.71	12.6
1.11	0.88	0.79	0.43	2.69	6.3
0.82	0.92	1.12	0.71	0.99	1.4
0.75	0.78	1.04	0.99	1.84	1.9
0.55	0.82	1.49	1.13	1.98	1.8
0.58	0.54	0.93	0.99	1.70	1.7
0.68	0.68	1.00	0.42	1.99	4.7
0.21	0.17	0.80	0.29	0.98	3.4
0.26	0.16	0.60	0.57	1.27	2.2
0.50	0.08	0.16	0.43	4.54	10.5
0.68	0.06	0.09	0.43	2.85	6.6
0.61	0.30	0.49	0.71	3.12	4.4
0.44	0.31	0.70	0.29	1.81	6.2
0.43	0.51	1.20	0.43	0.14	0.3
0.57	0.50	*0.88*	0.57	1.55	*2.7*
		AVG. = 0.74			AVG. = 7.3
		$n = 60$			$n = 60$
		SD = 0.26			SD = 7.5
		Overestimates by 35%			Underestimates by 86%

to 100 ml, as above. These samples were then Protargol silver stained (Lee et al., 1985) for detailed ciliate species identification and creation of a permanent slide collection of encountered protist species.

Macrozooplankton & larval fish ampling and analyses

Samplings for zooplankton and larval fish were carried out via quantitative, oblique plankton net tows. Two mesh sizes of 0.5 m diameter nitex nets (202 μm and 500 μm) and one 1000 μm 1 m diameter net were used. Each net was fitted with cod ends possessing windows with the same mesh sizes as the net's, as well as with two flow meters. One flowmeter was mounted within the net's brass ring mouth, the other outside the mouth of the net. This dual meter system allows for accurate estimation of the volume of water sampled (UNESCO, 1978). The 0.5 m net tows were carried out obliquely from bottom water to surface, with slow, continuous retrieval of hydrowire cable over several minutes while the ship was under way at about 1 knot. The 1 m net tow for larval fish collections was carried out by obliquely drawing the net through the water column, allowing it to fall to the bottom and again retrieving it 4 times, while under way at 3 knots.

The 202-μm mesh net was used to capture the copepods. That mesh size trapped adult copepods and advanced juveniles (copepodites), but early stages (nauplii) likely escaped due to their small size. For all net sampling, we endeavored to maintain a constant retrieval rate, and a constant angle between the wire and sea surface in order to integrate the sample over depth. All tow-collected net cod-end contents were placed with some seawater into glass jars containing 100 ml of 10% formalin buffered with phosphate. The final concentration varied between approximately 1 and 5%.

Clogging was evaluated from the ratio of the inside flowmeter to the outside flowmeter, which provided a rough estimate of filtration efficiency. This is considered an acceptable field check, with the caveat that the outside flowmeter may be biased by water accelerating out and around the net if it begins to clog (UNESCO, 1968). An efficiency of 1.0 would imply that the net did nothing to block the flow of water. Filtration efficiencies above 0.85 are the standard set by UNESCO (1968) to assure the accuracy of volume filtered estimates.

Net tows were replicated (repeated) on thirteen occasions to test for local (intrastation) patchiness. Lloyd's (1967) index of patchiness was used to measure the intensity of patchiness, and Fisher's index was used to test for statistical significance.

Prior to examination, copepod samples were removed from their preservative with a 200-μm sieve and placed in filtered LIS water. The concentration of organisms was adjusted by appropriate use of dilution and a plankton splitter, until there were approximately 100 – 300 copepods in a 5-ml subsample of the resulting reference volume (UNESCO, 1968; Omori & Ikeda, 1984). Three 5-ml analytical subsamples were taken from the reference volume with a Stempel pipette and placed into examination dishes with raised grids (Omori & Ikeda, 1984). Examination took place on an image analysis system consisting of an Olympus Model SZHILLD 7.5 – 128 power dissecting microscope and a video system linked through an IBM type personal computer with Olympus Cue 2 software. The coefficient of variation was calculated for each set of three subsamples as a measure of counting error (Snedecor & Cochran, 1967).

Copepod biomass estimates

Copepod individual weight varies by species, temperature, nutritional history, and life stage, Due to variable animal weight loss caused by variability in the final concentration of preservative relative to retrieved sample volume, we could not use drying and weighing of the copepods as our method of estimating biomass. Biomass measurements using preserved samples can succeed only when preservative concentration and pH are kept within precise limits. If this is done, weight loss should regularly fall in the vicinity of 30% (Bøttger & Schnack, 1986). As an alternative we used published regressions of prosome length to weight for the three numerically dominant copepods, *Temora longicornis, Acartia hudsonica*, and *Acartia tonsa,* to estimate biomass. This procedure would account for the bulk of the annual population, and include the heaviest common copepod, *Temora longicornis.* Length is not changed by preservation, and can explain $\geq$94% of the weight variation found in individual samples (Durbin & Durbin, 1978; Dam, 1989). The regressions we chose gave unpreserved dry weight (UDW) as follows:

1. *Temora longicornis*: $\log(\mathrm{UDW}) = 3.064 * \log(\mathrm{Length}) - 7.6958$ (Klein-Breteler & Gonzalez, 1988)
2. *Acartia tonsa*: $\mathrm{UDW} = 19.56 * \mathrm{Length}^{3.955}$ (Durbin et al., 1983)
3. *Acartia hudsonica*: $\mathrm{UDW} = 20.74 * \mathrm{Length}^{3.724}$ (Durbin et al., 1992)

Length measurements were done on the image analysis system using a mouse-driven micrometer at the time that the counts were done. Fifteen individuals per species were measured in order of encounter in each of the counting subsamples, yielding 45 individual weights for each species. The average weight for each species was multiplied by its abundance to obtain its UDW biomass in units of μg m^{-3}.

Results

Physical parameters

The seasonal range of water temperatures was 0–27 °C (Figs 2 and 3). The cycle was analogous to Riley's (1956) data, and occurred in a synchronous manner at each station, except that Stamford led the other stations very slightly at times when temperature was undergoing rapid seasonal change. Transitory surface-bottom temperature gradients of 1–3 °C were noted during spring warming, but the water column was nearly isothermal for most of the summer. This suggests that the seasonal thermocline did not extend shoreward of the 10 m isobath. The water column inshore of the 10 m isobath would be expected to stay relatively mixed because of the turbulent interaction of wind and tidal energy with bottom friction

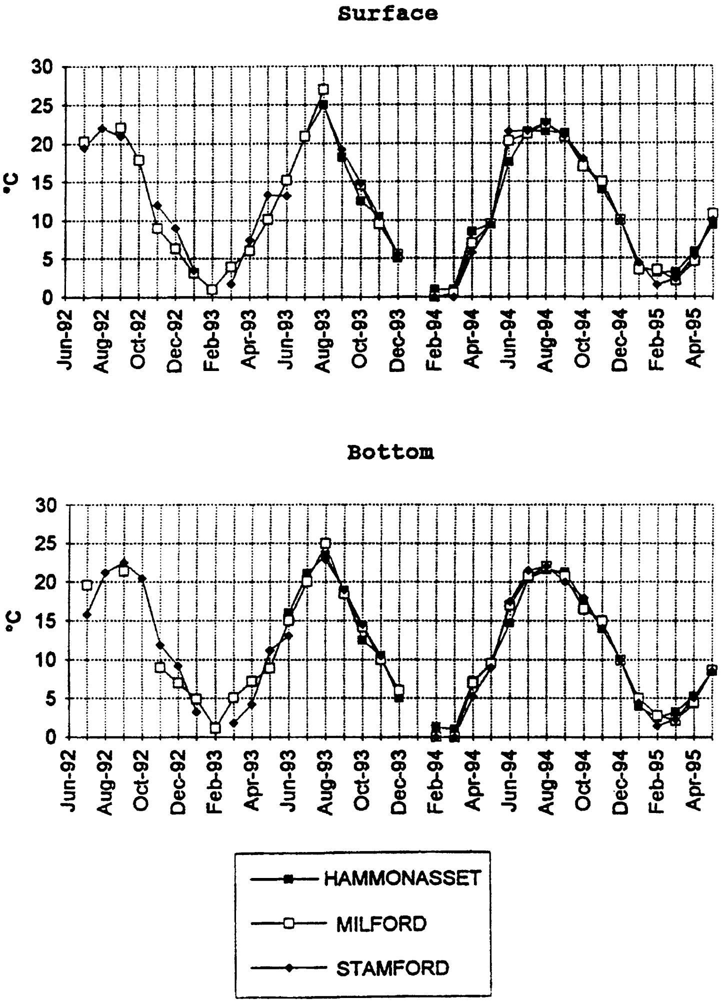

Figure 2. Annual Cycle of water temperature (°C) in the surface and bottom waters of our study sites.

and coastal features such as promontories and embayments, whereas the summer thermocline above the deep basins of LIS is typically somewhat less than 10 m deep (6–8 m) in the summer (Welsh & Eller, 1991; Johnson, 1993).

Most of the salinity data fell within 24–28 PSU, just above the center of the overall range of 20–30 PSU (Figs 4 and 5). Distinct seasonal cycles of spring lows and fall highs in salinity occurred at all stations, which Riley (1956) attributed to seasonal variation in snow-melt and run-off. The Hammonasset and Milford stations are within about thirteen and eight kilometers of river mouths, respectively, and both had broader ranges and greater variability of salinity than did Stamford. Increases between surface and bottom in average salinity were small (1–2 PSU), however, gradients over depth of several PSU occurred at times, particularly at the Hammonasset and Milford stations. There was a 1.3 PSU decrease in mean salinity between Hammonasset and Stamford, reflecting an overall east-west gradient that has been noted in LIS (Riley, 1956). The STD salinity data may have been up to 0.2 PSU in error on the low side based on a calibration of the instrument performed by the manufacturer since this study was completed.

Water column stability, as indicated by Δ sigma-t (Figs 6 and 7), depended primarily on salinity gradients rather than temperature gradients as shown by the correspondence between Δ sigma-t and Δ salinity (Fig. 6). On several occasions high values of Δ sigma-t (and Δ salinity) were noted at Hammonasset and Milford, perhaps due to their proximity to river

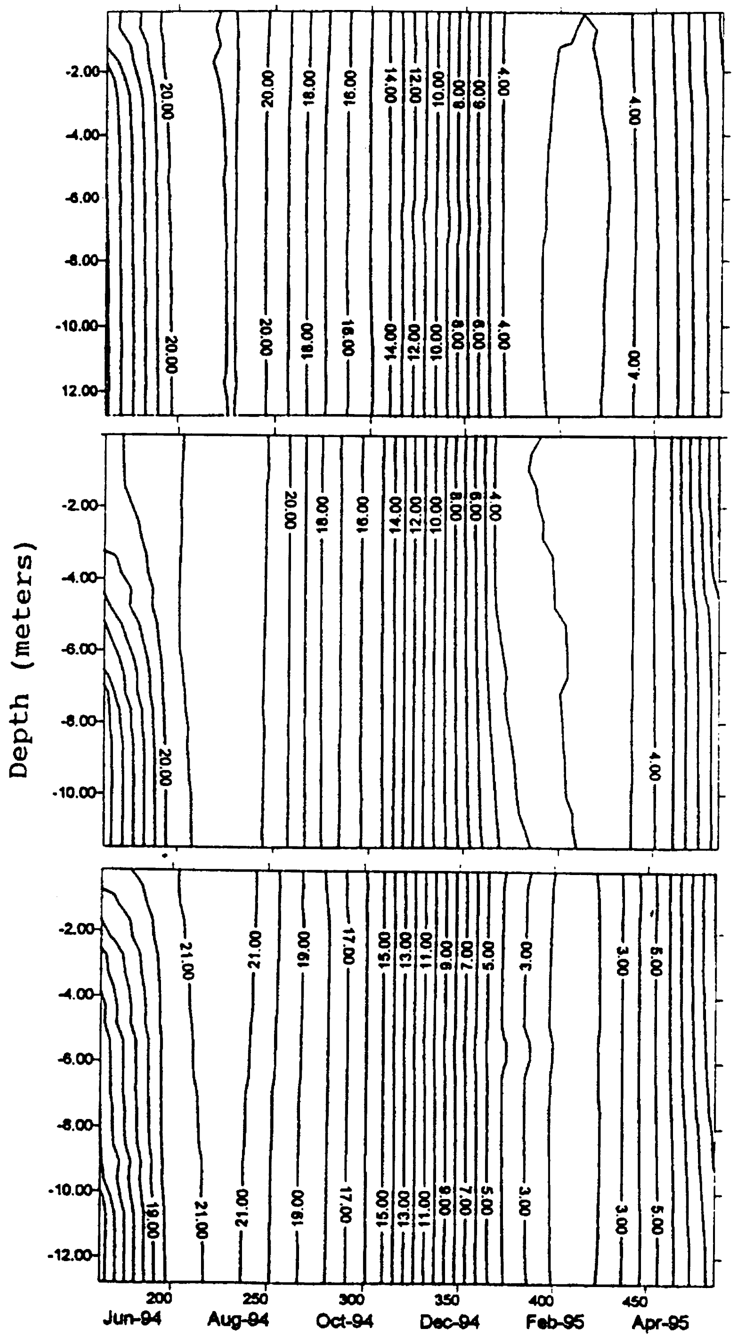

Figure 3. Vertical Contours of temperature (°C) at the Hammonasset (top), Milford (center) and Stamford (bottom) stations.

outlets. Stamford was the least variable of the three stations. All stations were stratified in June of 1993 and 1994, but they did not remain so over the summer. The water column appeared unstable (negative Δ sigma-t) on occasion, more often at Milford than at the other stations. The Δ sigma-t record obtained from STD profiles (June 1994–May 1995) was less variable than that obtained from prior measurements using a refractive salinometer and a mercury thermometer. While this difference could reflect real interyear variation, it is more than likely an artifact of the switch to the STD profiling unit. The temperature, salinity,

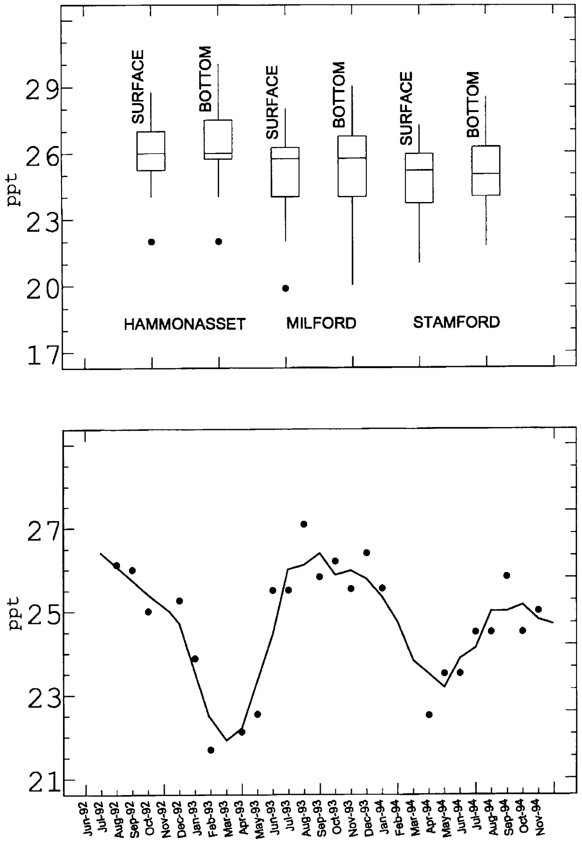

Figure 4. Box and Whisker Graph (top), shows the median salinity as a horizontal line, the central 50% of the data as a box, and the range as a vertical line. Below is an example from Stamford surface water of the seasonal salinity cycle. The solid line is a 3 point moving average. Values are in parts per thousand (PPT).

and Δ sigma-t data suggest that the mixing regime was broadly comparable among stations, but we suspect that the more frequent presence of a halocline at Hammonasset and Milford, and the westward decrease in tidal energy (Riley, 1956) probably led to differences at times.

In summary, these stations displayed similar seasonal variations of temperature, salinity, and Δ sigma-t, and were comparable to Riley's (1956) observations at near-shore stations. The Stamford station exhibited somewhat narrower ranges of salinity and Δ sigma-t than the other two stations, which are nearer large sources of riverine fresh-water. The persistent seasonal thermocline, which forms in the deeper portions of LIS, was absent.

The distribution of dissolved oxygen over depth (Fig. 8) was rather uniform and typically 80–100% saturated. There was a mild oxygen defecit during the summer and fall, which increased from east to west. Bottom hypoxia was not observed at our nearshore stations, during the study period.

Table 2. Correlations between surface and bottom water values of nitrogen. All 3 stations were grouped for the test

Parameter	*r*	*p*	n
Total dissolved nitrogen	0.75	<0.01	57
Particulate nitrogen	0.60	<0.01	45
Nitrate+nitrite	0.97	<0.01	65
Ammonium	0.53	<0.01	65
Dissolved inorganic nitrogen	0.90	<0.01	65
Dissolved organic nitrogen	0.67	<0.01	57
Total nitrogen	0.48	<0.01	45

Nutrients

Nitrogen results are provided in nine categories: total dissolved nitrogen (TDN), particulate nitrogen (PN), nitrate, nitrite, nitrate+nitrite (NO_x), ammonium (NH_3), dissolved inorganic nitrogen (DIN), dissolved organic nitrogen (DON), and total nitrogen (TN) (Figs 9–18). Strong seasonal cycles in NO_x and DIN were concurrent in the horizontal (east–west) axis and the vertical (surface–bottom) axis. Changes in the other nitrogen parameters, although not always so synchronized, generally shared the same seasonal time frame at each station and depth. Surface and bottom concentration values were significantly correlated with one another in all categories (Table 2). Station-to-station differences were clearest when concentrations were above average, and less clear, or perhaps too small to detect, when concentrations were below average.

Dissolved inorganic nitrogen (NO_3, NO_2, NH_4)

Nitrate

When chemical species were considered individually, Milford waters were found to have higher average nitrate concentrations than Hammonasset waters, and concentrations in Stamford waters were higher than those of both Milford and Hammonasset (Table 3). As expected, nitrate levels were fall/winter seasonally highest. As was true for the physical parameters, bottom and surface water nitrate profiles were similar to each other at all 3 stations. Nitrate patterns and highs were similar to those observed by Riley et al. in the 1950s for their near shore stations. Additionally, significant and meaningfull year to year variability was observed in nitrate patterns at our stations (Fig. 9).

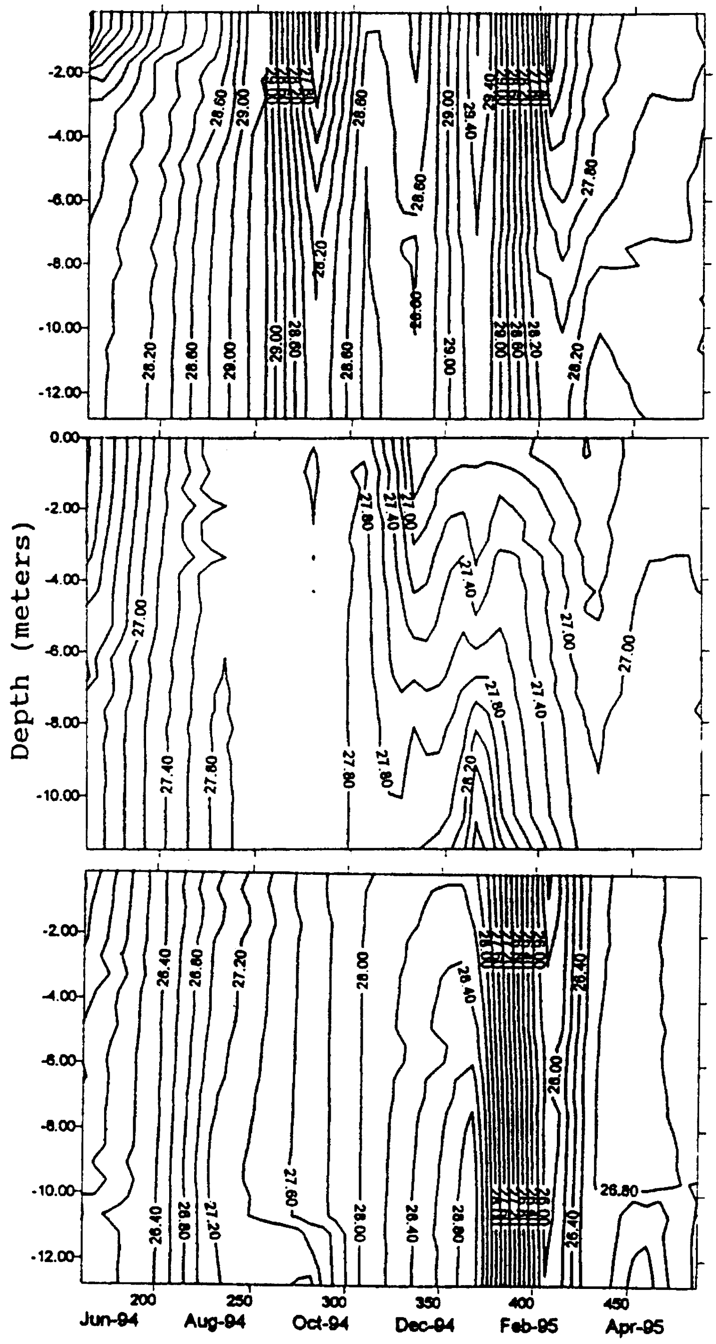

Figure 5. Vertical Contours of salinity (Parts per thousand, PPT) at the Hammonasset (top), Milford (middle) and Stamford (bottom) station.

Nitrite

Nitrite levels were consistently low (mostly in the 1 μg-atm range with occassional highs of 4 μg-atm, Figs 9 and 10) with minor peaks corresponding to highs in the nitrate fall/winter concentrations.

Ammonium

The range of NH_4 concentrations was from the detection limit (~0.1 μM) to 10 μM (Figs 9 and 10). Ammonium ion concentrations (after the normal seasonal variations were accounted for) were far more variable than nitrate and nitrite levels, although, over-

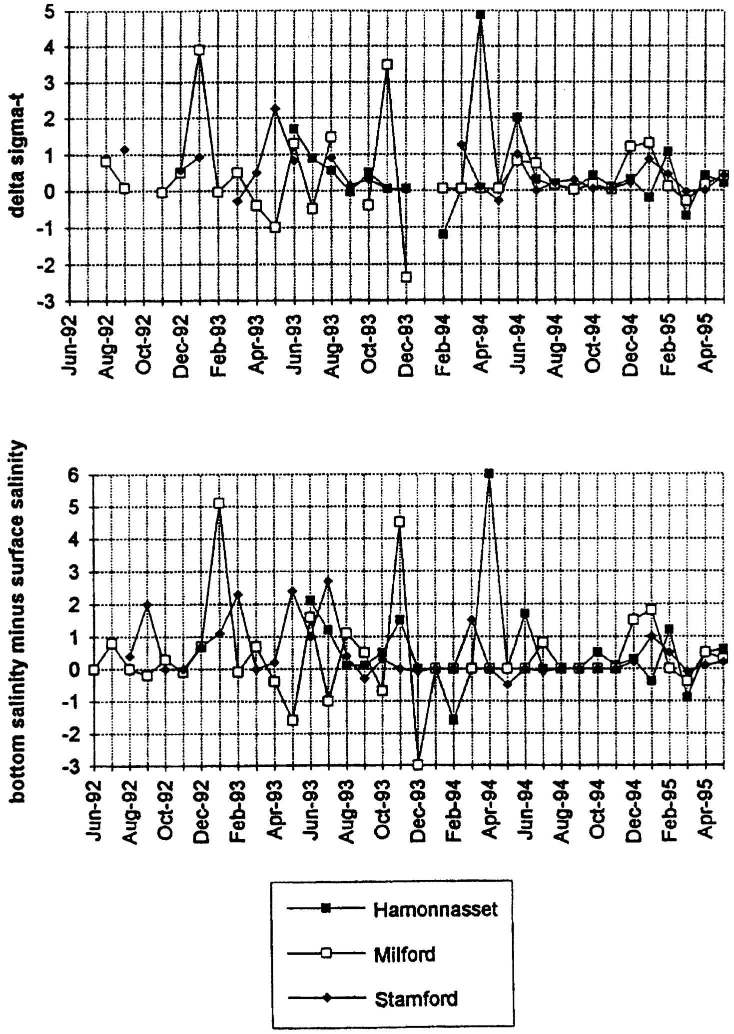

Figure 6. Water column stability expressed in sigma T density units, or (kg/m^3) -1000 (top) and bottom salinity minus surface salinity in PPT (bottom).

all they tended to follow NO$_X$ patterns. Seasonal trends at the different stations seemed to be out of phase with each other from June 1992 until April 1993. The pattern between stations in 1994 and 1995 was more coherent. Values tended to be above average in August 1993, December 1993, April 1994, and August–September 1994. No one station had consistently lowest NH$_3$, and station-to-station differences produced no overall trend in mean concentration at the surface over the period of study. Bottom water concentrations were higher than surface levels likely due to the influences of microbial and animal metabolic activities. At the bottom, concentrations decreased between Hammonasset and Milford, and increased between Milford and Stamford, with Stamford highest overall (Table 3). Stamford waters exhibited greater differences and higher peaks than the other 2 stations. The mean NH$_3$ concentration in bottom water was higher than at the surface (Table 3), and there were strong contrasts between surface and bottom some months (note June-September 1993 at Stamford).

Nitrate + Nitrite

The range of NO$_X$ concentrations was from below the detection limit ($\sim$0.1 μM) to 28 μM (Fig. 11). There was an obvious seasonal-cycle with values above average from September or October until February or March, and below average from February or March until August or September. In fall 1992, NO$_X$ was more abundant than in fall of the two following years,

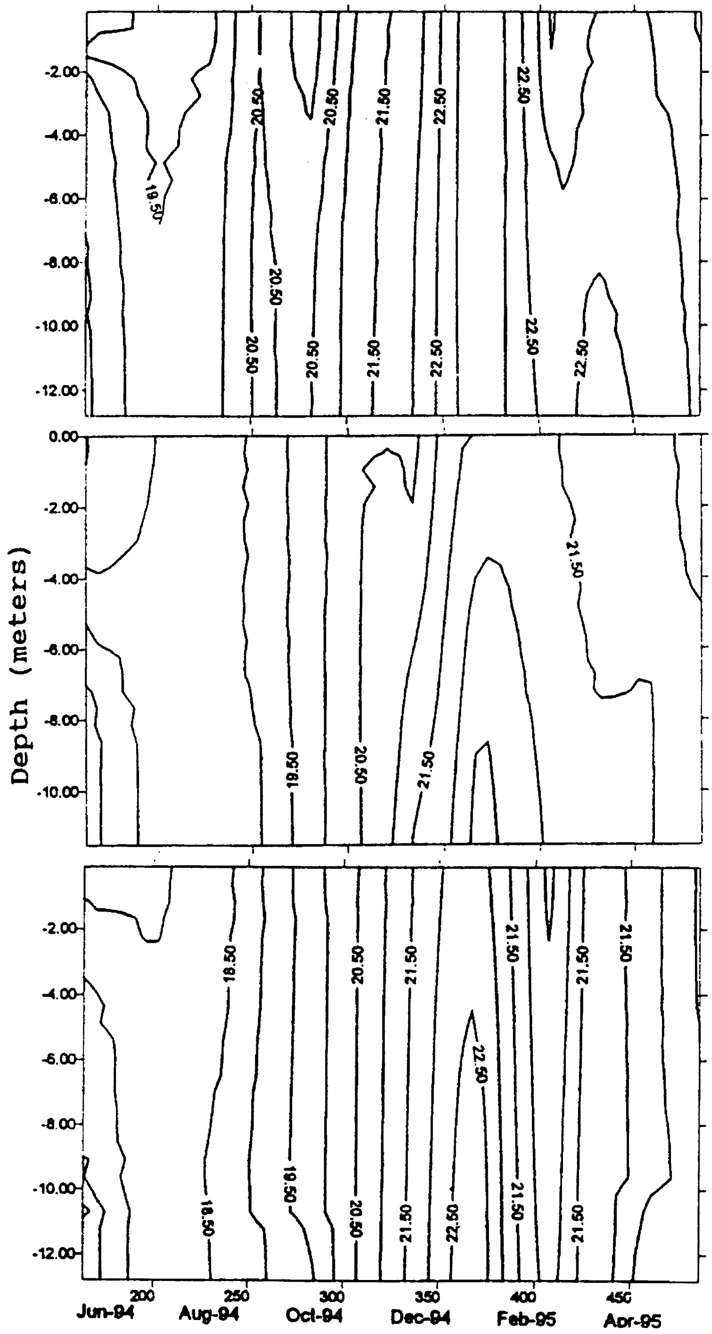

Figure 7. Vertical Contours of density expressed in sigma T units, or (kg/m^3) −1000 at Hammonasset (top), Milford (middle) and Stamford (bottom) stations.

which opposes the year-to-year pattern in Chl. No one station had lowest NO_x at all times, but Hammonasset was usually lowest during the fall-winter peak. Moreover, station-to-station differences were consistent enough to produce modest east–west trends of increase in mean concentration over the period of study (Table 4). Average surface concentrations increased between Milford and Stamford, and bottom concentrations increased between all three stations. The mean concentration was slightly higher at the

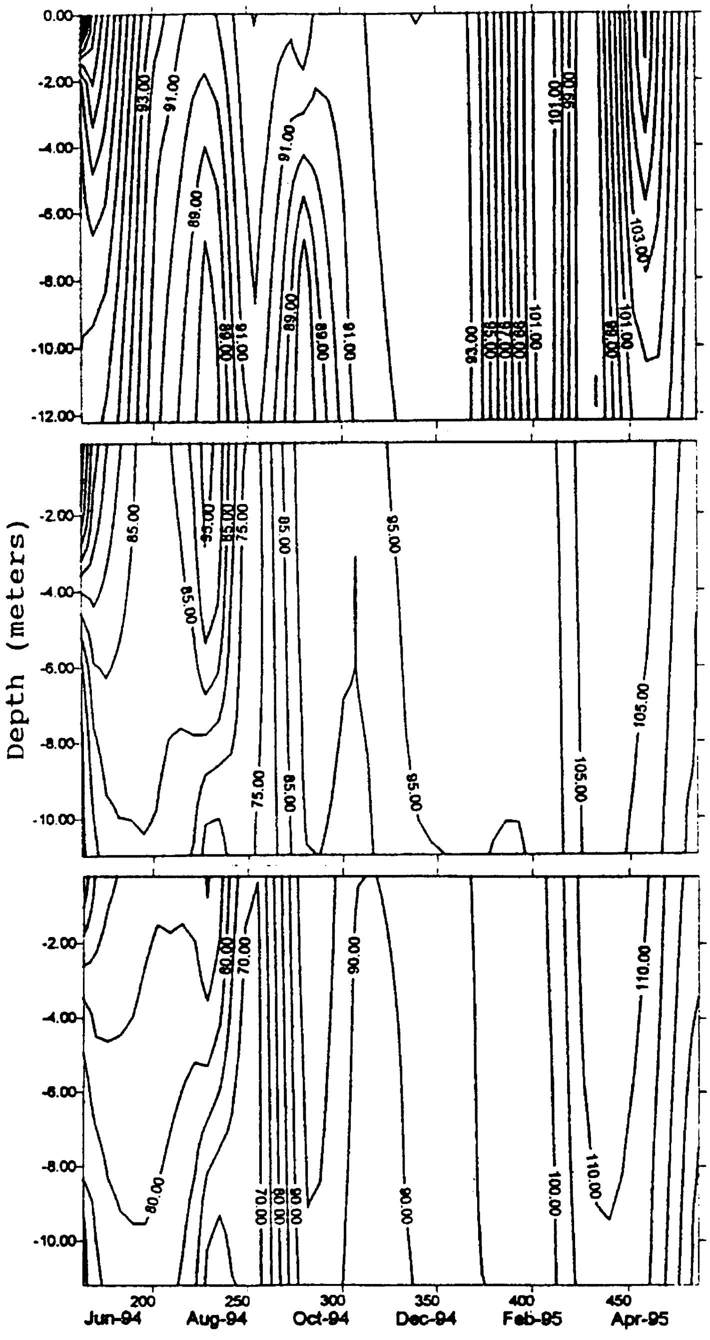

Figure 8. Vertical Contours of dissolved oxygen in units of % saturation, at the Hammonasset (top), Milford (middle) and Stamford (bottom) stations.

surface at Hammonasset, and slightly higher at the bottom at Milford and Stamford (Table 4).

Dissolved inorganic nitrogen

The range of DIN (NO_X plus NH_3) concentrations was from 0.13 μM to 29 μM (Fig. 12). On average, most (75%) of DIN was in the form of NO_X, thus the strong positive correlation between temporal variation in DIN and NO_X (Tables 3 and 4) is largely autocorrelation. Concentrations were high from September until February, and low from March through August. There was a clear pattern with Hammonasset lowest when DIN

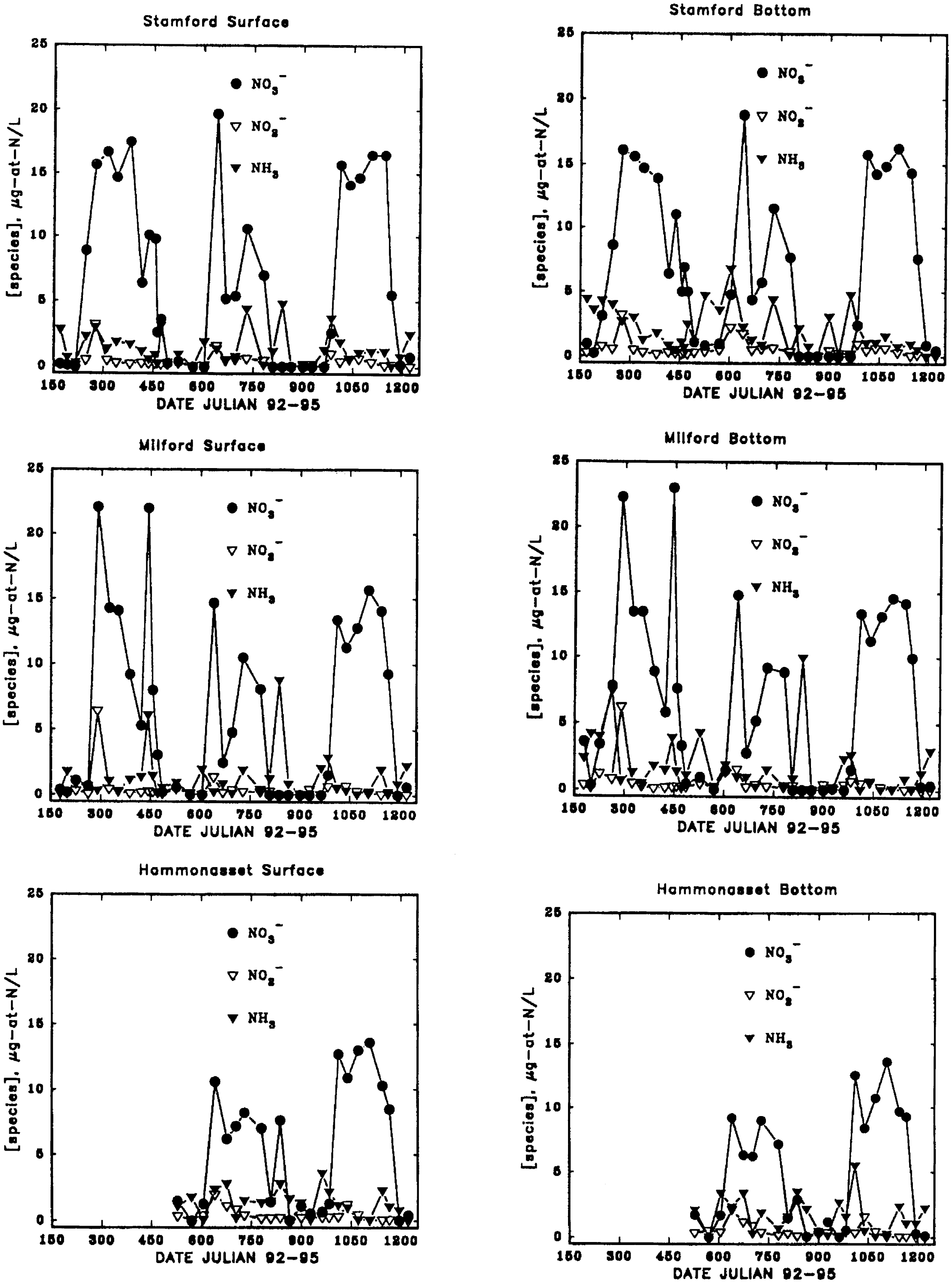

Figure 9. Concentrations of nitrate, nitrite and ammonia in surface and bottom waters over the course of this study. Dates are presented as Julian values beginning January 1, 1992.

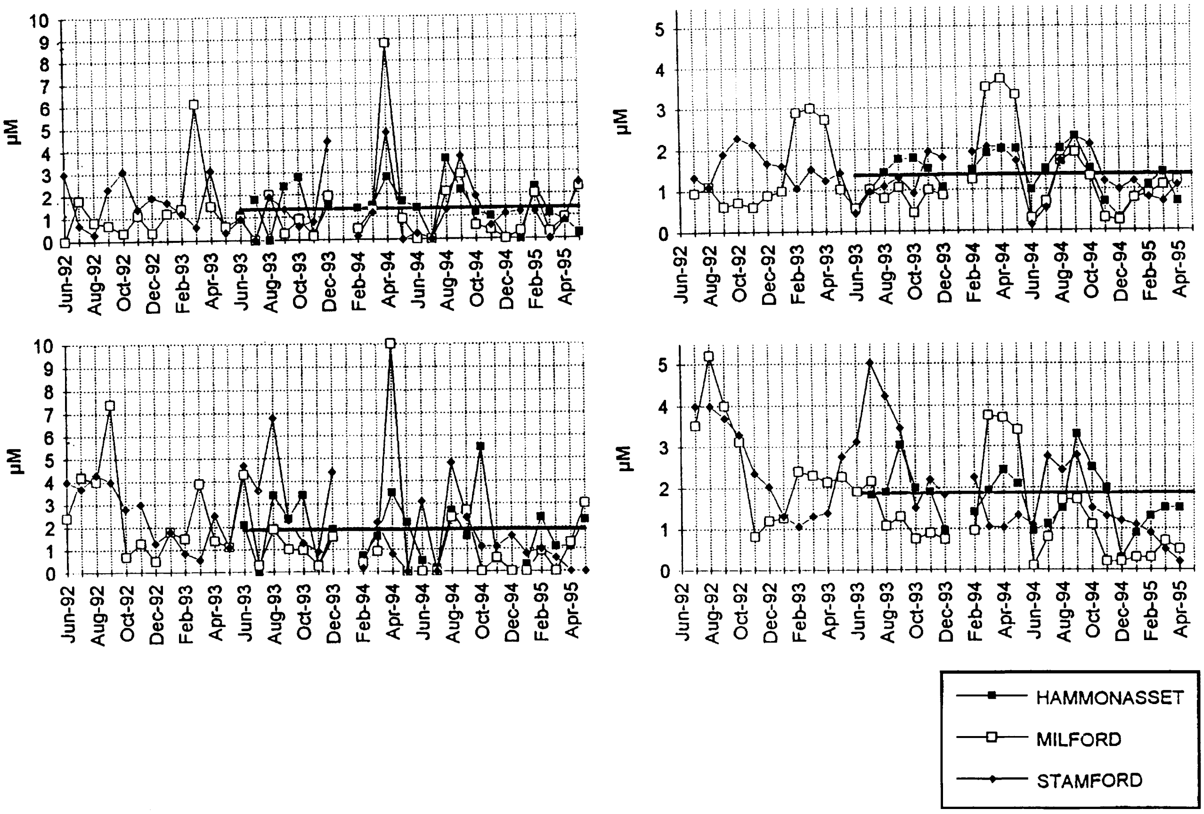

Figure 10. Ammonia concentrations in surface (above) and bottom (below) waters of the 3 study sites. On the right, data are presented as 3 point running averages. The straight lines are the composte means for the 3 stations for the 2 year period when all 3 stations were sampled.

was high in the fall and winter of 1994–1995. No one station had consistently lowest DIN, and station-to-station differences produced no overall trends in mean concentration over the study period between Hammonasset and Milford. Stamford, however, exhibited a small but consistently higher TIN level at the surface, and a larger one at the bottom (Tables 3 and 4). The overall mean for bottom water was slightly greater than for surface water at Milford and Stamford, but not at Hammonasset (Tables 3 and 4).

Total dissolved nitrogen

The range of concentrations of total dissolved nitrogen was 7.6 – 40.2-μM (Fig.13). TDN was commonly above average from September until February, and below average from April until August. No one station had lowest TDN all the time, but Hammonasset often did (Fig. 13), and these station-to-station differences were consistent enough to produce modest east-west trends of increase in mean concentration over the period of study (Table 4). The increase occurred between Hammonasset and Milford at the surface, and between all three stations at the bottom. The TDN mean was slightly higher at the surface than at the bottom at each of the stations (Table 4).

Dissolved organic nitrogen

The range of DON (TDN minus DIN) concentrations was from 0.8 to 28.2 μM (Fig. 14) however, only one value was below 4.4 μM (September 1993 at Hammonasset). Temporal variation was often (but not always) coherent between stations and between depths, and the patterns were not as distinct or regular as for DIN. Values were below average in March and April 1994, and again for an extended period from November 1994 through April 1995. Values were above average in October and November 1993, and again in June and July 1994. The October–November high of 1993 was not repeated in 1994. No one station had consistently lowest DON, but station-to-station differences were consistent enough to produce modest overall trends in mean concentration over the period

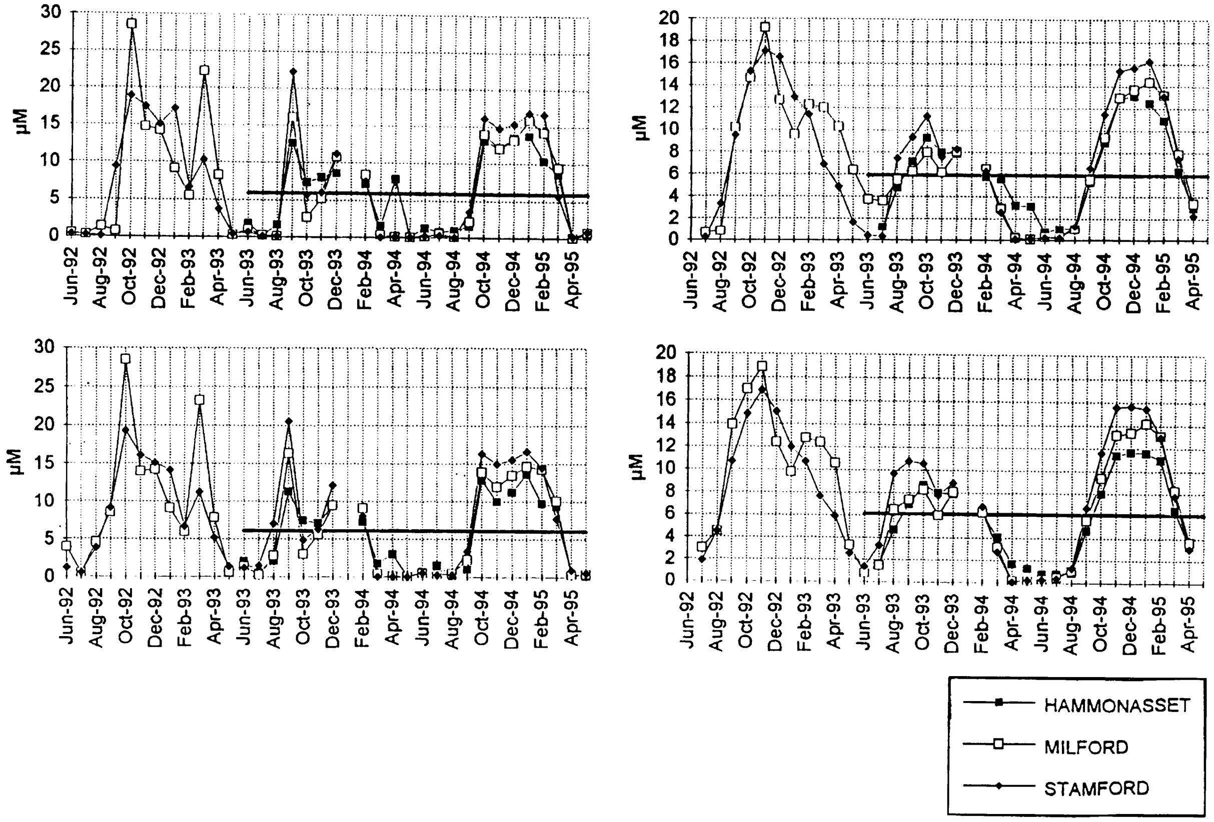

Figure 11. Nitrate and nitrite (NO_X) concentrations in the surface (above) and bottom (below) at the 3 study sites. See Figure 10 for data presentation details.

of study (Table 4). On average, there was an increase between Hammonasset and Milford at the surface, and small east–west increases between all three stations at the bottom. There were differences between surface and bottom at times, but with no clear pattern. Overall, there was slightly less DON in bottom-water relative to the surface (Table 4).

Particulate organic nitrogen

Particulate organic nitrogen (PON) at the three sites varied generally between 2 and 42 μM, but only one measurement (August 1994 at Hammonasset) exceeded 28 μM (Figs 15–17). Seasonal cycles in PON are not pronounced, although slight increases in early spring and late summer may be discerned. With the exception of a peak in Hammonasset during August, 1994, large accumulation of nitrogen in standing stocks of suspended microbial biomass does not appear to be occurring. PON, summed for the entire study, is only slightly higher in Stamford than in Milford and Hammonassett; the difference is not statistically significant (ANOVA $p>0.05$). Peaks of PON corresponded with phytoplankton blooms that took place in August, October, and March. There were tendencies for PON to be below average when TDN was above average, from October to December, for example in 1994. No one station had consistently lowest PON, and station-to-station differences were consistent enough to produce only small east–west trends of increase in mean concentration over the period of study (Table 4). These small increases occurred between all three stations at the surface, and between Milford and Stamford at the bottom. There were sometimes distinct differences between surface and bottom, coincident with surface-bottom chlorophyll differences: bottom-PON exceeded surface-PON in March 1994, and vice-versa in August 1994. There was an overall tendency for PON in bottom waters to be slightly higher than at the surface (Table 4).

It should be noted that PON represents N contained in photosynthetic and heterotrophic microorganisms, as well as non-living particles. Factors limiting accumulation of PON include nutrient uptake

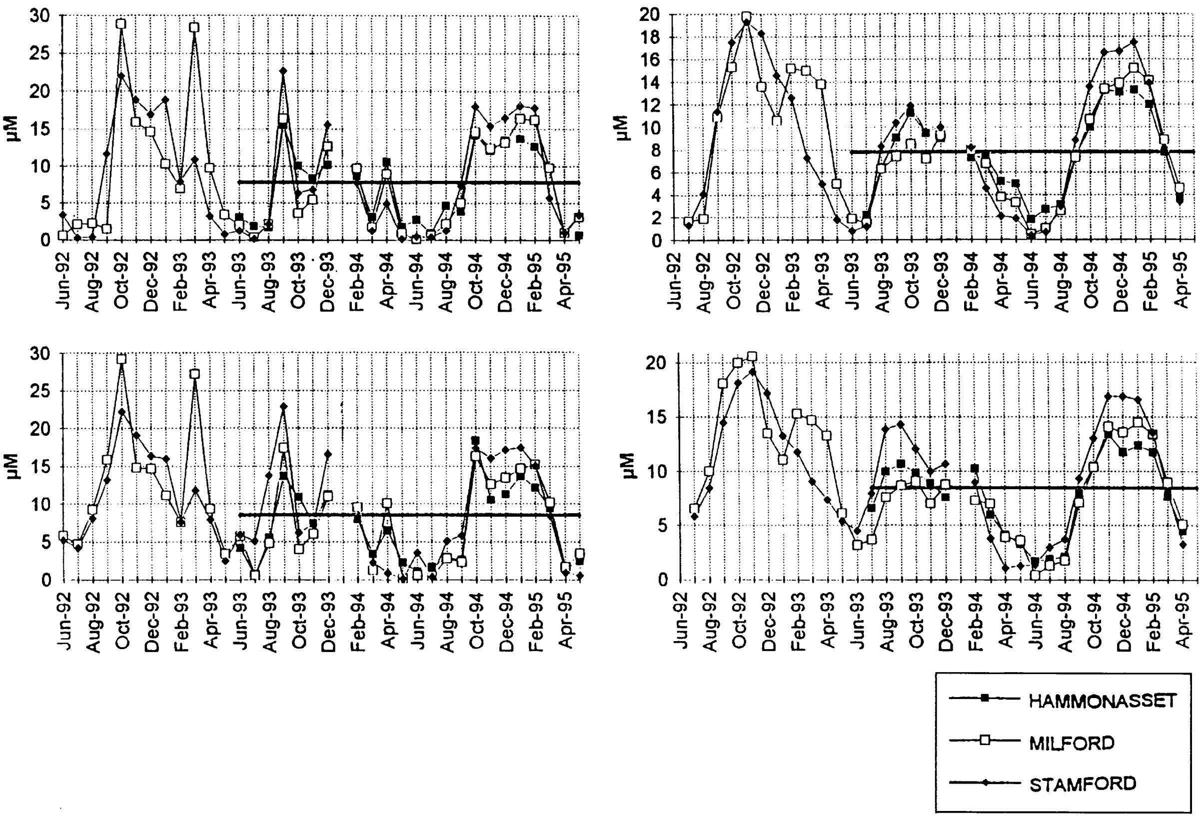

Figure 12. Dissolved inorganic nitrogen (DIN) concentrations in the surface (above) and bottom (below) waters of the 3 study sites. See Figure 10 for data presentation details.

by microplankton, consumption of microplankton by meioplankton, and sinking, either as whole cells or as fecal pellets. Some evidence that sinking was an important sink for PON during this study is found in consistently higher PON values in bottom samples than in surface, when values are averaged for the entire study (Fig. 18). Interestingly, differences between surface and bottom PON values are more pronounced in Hammonasset and Stamford than in Milford. Microscopic observations suggest that PON in Hammonasset bottom samples may be accounted for to a large extent by benthic diatoms; whereas, bottom-water PON in Stamford is likely dominated by zooplankton fecal pellets.

Total nitrogen

The range of TN (TDN plus PON) concentrations was from 17.1 to 61.5 μM (Fig. 18), but only one value exceeded 50 μM (August 1994 at Hammonasset). Temporal patterns consisted of decreases and below average values from February until May in 1994, and above average values from September until November in 1993 and in July and August in 1994. No one station had continually lowest TN, and station-to-station differences were consistent enough to produce only small trends in mean concentration over the period of study (Table 4). On average, there was an increase between Hammonasset and Milford at the surface, and an increase between Milford and Stamford at the bottom. Differences between depths were obvious at certain times (note fall 1993 at Milford, and August 1994 at Hammonasset).

Nitrogen summary

The distribution of nitrogen compounds varied seasonally; the proportion of DIN in TN ranged below 1% during spring and summer, to as high as 60% during fall and winter; the proportion of DON in TN ranged around 50–60% in summer and fall, but could drop in winter to 30–40%; the proportion of PN in TN averaged 35%, but ranged as high as 70% during blooms. These seasonal variations took place in a more-or-less concurrent manner at each station, consistent with the behavior expected of nitrogen in a temperate estuary

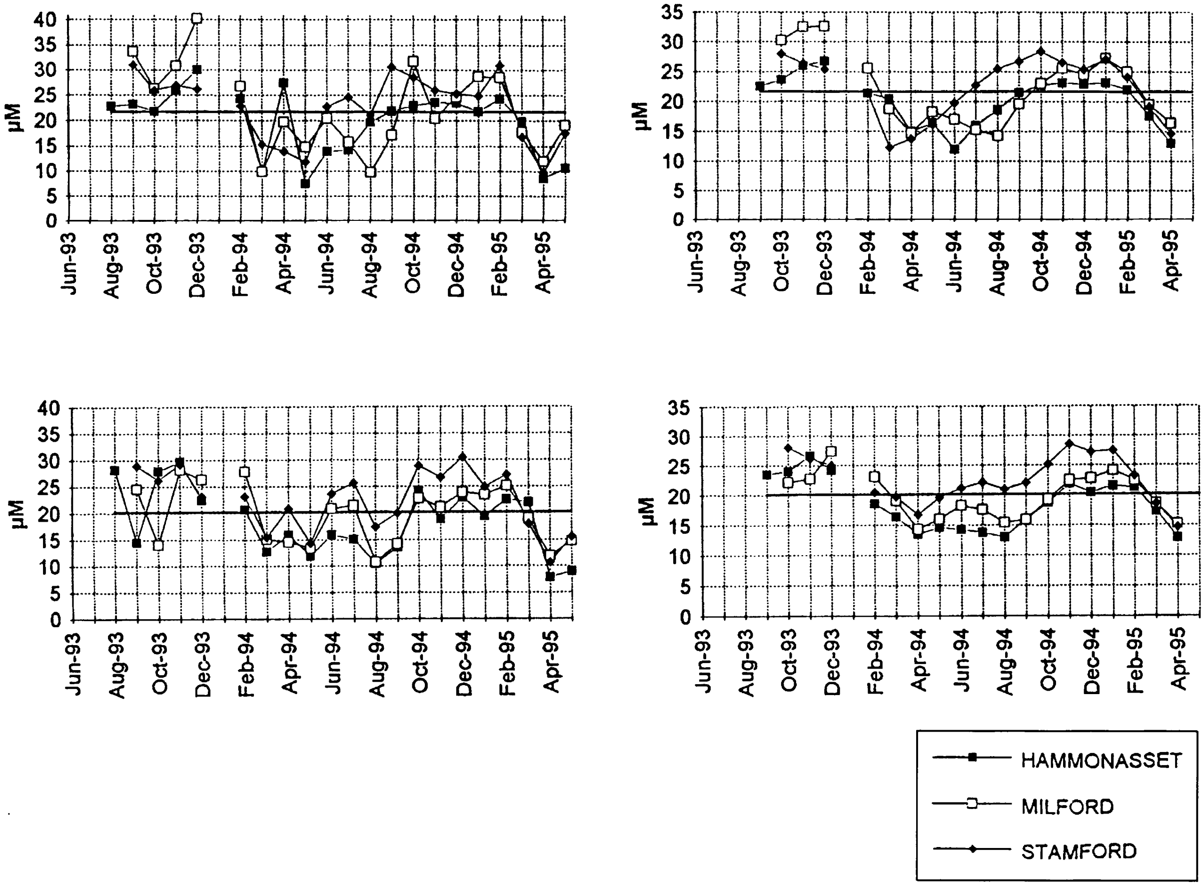

Figure 13. Total dissolved nitrogen (TDN) concentrations in surface (above) and bottom (below) waters, of the 3 study sites. See Figure 10 for data presentation details.

such as LIS (Harris, 1959). In the midst of the seasonal changes, there emerged trends of east–west increase in TN, TDN, DON, PON, DIN, NO_x, and NH_3 for the period of study as a whole. Although they were small (10–20%), these trends may be meaningful because they cover 2 years, appeared in every category, and are consistent with an increase in eutrophication from east to west in LIS.

Inorganic phosphorous concentrations

Dissolved ortho-phosphate concentrations follow a well defined seasonal cycle of highs in the fall/winter and lows in the spring/summer. Levels were consistently higher in Stamford surface & bottom waters as compared with Hammonasset waters (Fig. 19 and Table 3). Differences were more pronounced in the bottom waters where concentrations tended to be higher overall, relative to surface waters. Milford concentrations were very similar to Stamford water levels throughout this study.

Dissolved inorganic N & total dissolved N to phosphorous ratios

The Redfield Ratio of 106:16:1 (C:N:P) is the starting point for discussing nutrient ratios important to planktonic organisms. This ratio represents an approximate ideal elemental ratio for phytoplankton and zooplankton. When N:P ratios drop below approximately 16:1, hindrances to balanced growth occur. In reality, the ideal ratio varies much by species. For this reason as deviations from ideal occur differential successes among species result. Nonetheless, on average, for phytoplankton significant decreases in this ratio favor microalgal forms that can supplement their diets heterotrophically (e.g. the dinoflagellates). Typically the Redfield Ratio is calculated based on dissolved inorganic nutrient levels. However, dissolved organic

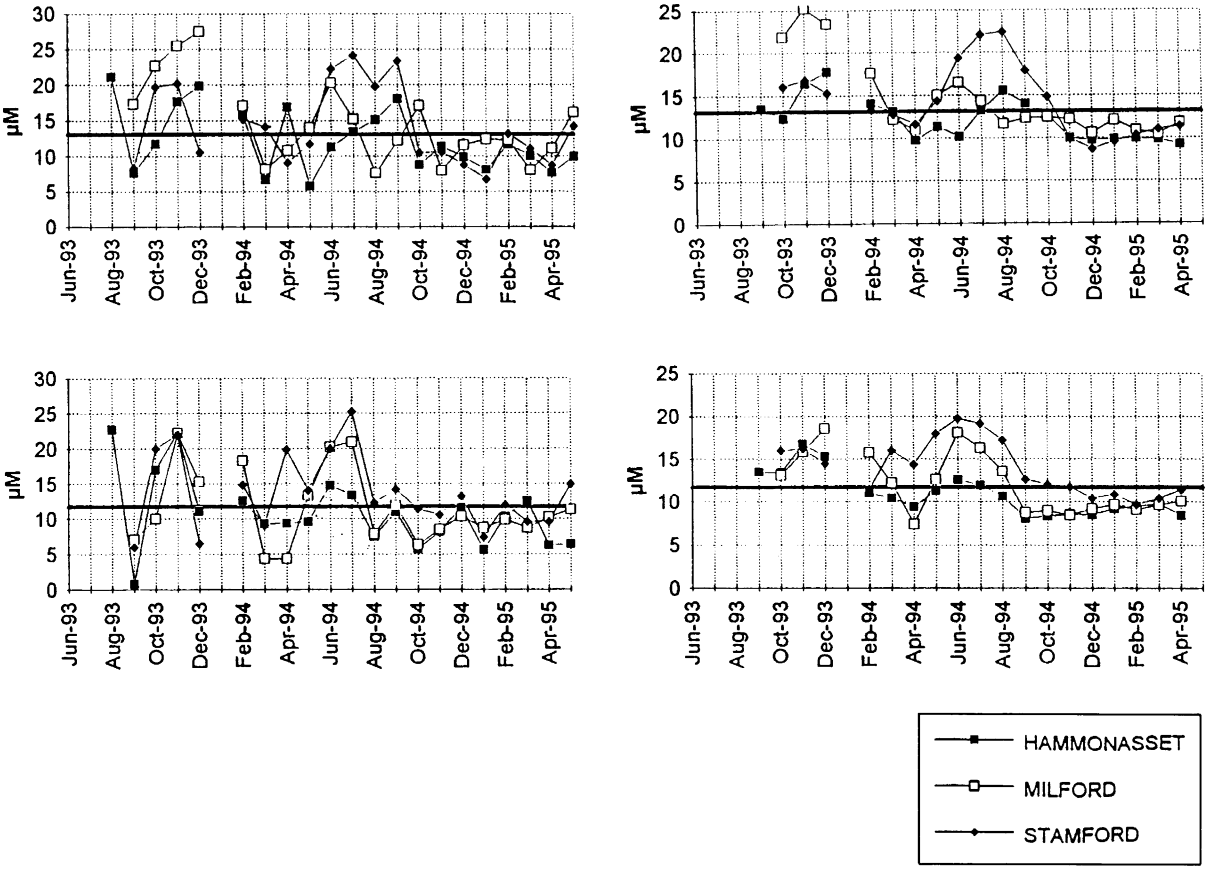

Figure 14. Dissolved organic nitrogen (DON) concentrations in surface (above) and bottom (below) waters, of the 3 study sites. See Figure 10 for data presentation details.

nitrogen (DON) is utilized as a nitrogen source by many (but not all) phytoplanktonic species. It is, therefore, important to calculate N:P ratios based on both inorganic and organic dissolved N levels, as we have done in this study (Tables 5 and 6). Ratios varied much over the year for Long Island Sound nearshore waters. Based on inorganic nutrients only, Stamford waters often exhibited water column values below 10, with an average of 5.6 and 5.8 for years 1 and 2 of this study. Corresponding values of 8.7 and 5.5 for Milford waters and 7.6 and 7.3 for Hammonasset waters were found. Respective bottom water values of 5.1, 7.1 and 6.3 (year 1) and 5.7, 5.4 and 7.2 (year 2) were found for Stamford, Milford and Hammonasset. When total dissolved nitrogen levels are considered, Stamford surface values of 32 and 21.8, Milford levels of 38.1 and 21.3 and Hammonasset levels of 21.8 and 24.5 were found, with respective bottom water values of 27.2 and 17.5 (Stamford), 24.6 and 16.1 (Milford) and 18.8 and 18.8 (Hammonasset) during this study.

Biologically active silica

Dissolved, biologically-available silica was measured because it is a required macro-nutrient for one group of phytoplankton, the diatoms, for incorporation into cell walls. The main sources of BSi in coastal seawater are runoff from shore (Si derived from Chemical weathering of crustal rock), and dissolution of silica minerals (chiefly old diatom shells) in bottom sediments. Dissolution rates of diatom shells are slow, so that little recycling occurs in the water column. Removal of BSi from solution can be attributed almost entirely to uptake by diatoms. Hence, the BSi content of a parcel of water represents the potential for that water to support diatom growth, assuming that all other algal nutrients are present as well.

The theory that nitrogen (N) limits phytoplankton production in Long Island Sound (LIS) is not inconsistent with the hypothesis that BSi limits diatom production. Indeed, human activities have been

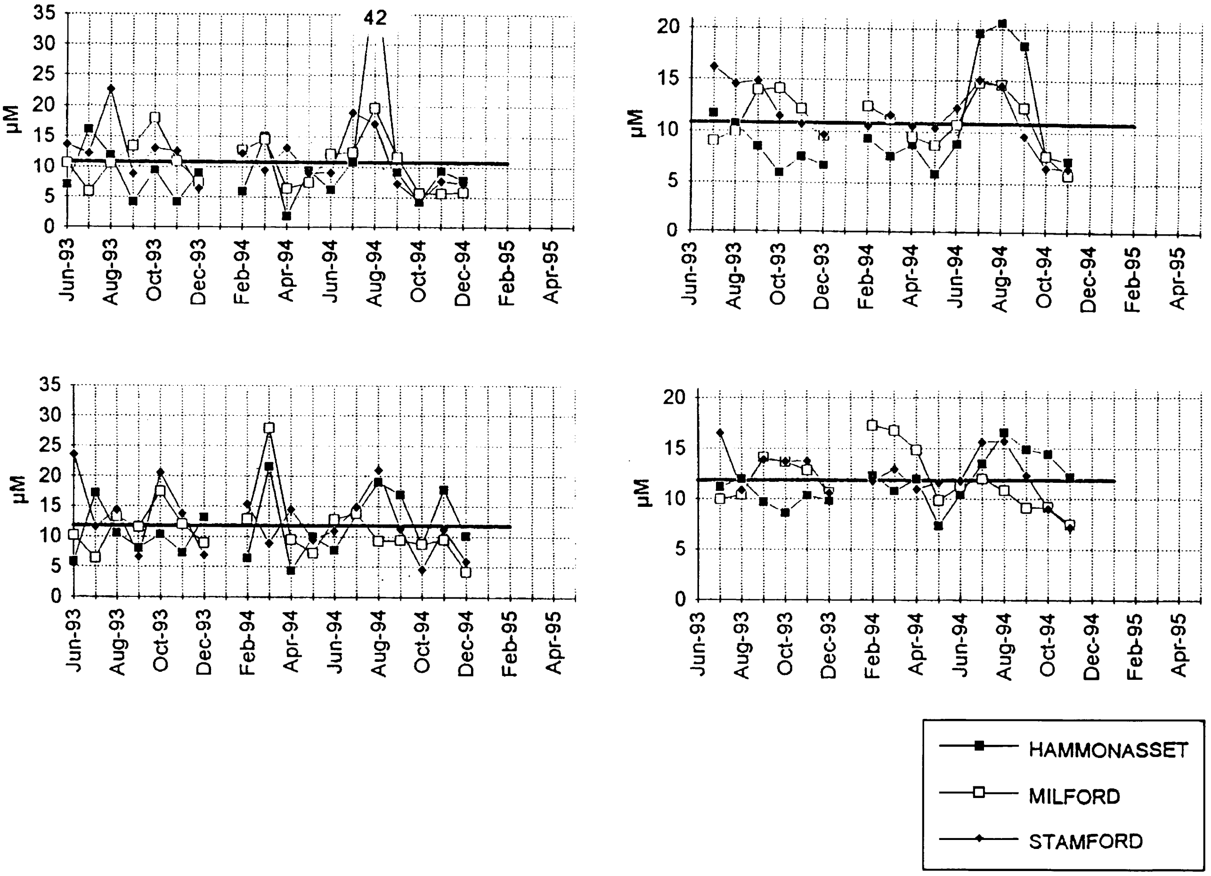

Figure 15. Particulate nitrogen (PN) concentrations in surface (above) and bottom (below) waters, of the 3 study sites. See Figure 10 for data presentation details.

shown to increase N inputs; whereas, we have done relatively little to increase BSi inputs into LIS. Hence, N over-enrichment has two potential effects upon phytoplankton production in LIS: (1) to increase total phytoplankton production, and (2) to increase the percentage of phytoplankton production attributable to non-diatom algal taxa. Whether primary production is in the form of diatoms or other algal cells can have consequences to higher trophic levels if diatoms are better (or worse) sources of food for certain herbivores. One algal taxonomic group that may grow in the absence of diatoms – the dinoflagellates – has the potential to disrupt pelagic food webs through production of toxic metabolites. Biologically available silica, therefore, plays a pivotal role in ecological consequences of N over-enrichment.

BSi concentrations in LIS seawater during this study ranged from <1 μM to approximately 60 μM (Figs 20–22), with one exceptionally high value of 180 μM in Stamford bottom waters in July of 1993. We attribute this high BSi value to enhanced dissolution of sediment Si during an hypoxic event, but cannot confirm this since we did not measure dissolved oxygen in 1993. The most noticeable variation in BSi occurred seasonally. During most of the summer of 1994, BSi values remained in the range of 5–10 μM. As a general rule of thumb, BSi concentrations between 5 and 2 μM become limiting to diatoms (Egge & Aksnes, 1992). Thus, it appears likely that diatoms may have been limited by BSi during the summer of 1994. The increase in BSi during autumn and winter is an expected consequence of light-limited diatom growth. Why BSi concentrations remained low during the winter of 1993–94 is somewhat puzzling, but this observation does demonstrate the range of inter-annual variability.

Although seasonal changes in BSi are clear, differences between sites are less obvious from a simple plot. Mean BSi values for the three sites, averaged for both surface and bottom waters for the entire study period, reveal slightly higher BSi in Stamford

Table 3. Summary of total nitrate, ammonia, inorganic nitrogen, and reactive phosphate concentrations for the 3 stations over the entire study period. Ammonia and phosphate values are presented as total water column averages, as well as separately for surface and bottom waters. All other values represent total water column averages

Average total nitrate (μg-at-N/L)

	92–93	93–94	94–95
Stamford	7.7	5.7	7.3
Milford	8.3	4.7	6.6
Hammonasset		4.8	5.8

Average total ammonia (μg-at-N/L)

		Stamford	*Milford*	*Hammonasset*
1992–1993				
	Surf.	1.5	1.3	
	Bot.	2.3	2.3	
Water column		1.9	1.8	
1993–1994				
	Surf.	1.3	0.90	1.4
	Bot.	2.9	1.3	1.8
Water column		2.1	1.1	1.6
1994–1995				
	Surf.	1.2	1.0	1.2
	Bot.	1.4	1.0	1.5
Water column		1.3	1.0	1.4

Average total inorganic nitrogen (μg-at-N/L)

	92–93	93–94	94–95
Stamford	10.4	8.8	8.9
Milford	11.9	6.5	7.9
Hammonasset		7.1	7.5

Average reactive phoshate (μg-at-P/L)

		Stamford	*Milford*	*Hammonasset*
1993–1994				
	Surf.	1.1	1.0	1.0
	Bot.	1.4	1.3	1.1
Water column		1.3	1.1	1.0
1994–1995				
	Surf.	1.3	1.3	0.92
	Bot.	1.4	1.4	1.0
Water column		1.3	1.3	0.96

and Milford than in Hammonasset (Figs 20–22), but this difference is not statistically significant (ANOVA $p>0.05$). The difference between surface and bottom waters, summed for the entire study, was statistically significant (ANOVA $p>0.05$), with bottom waters containing more BSi. It is interesting to note that the difference between surface and bottom BSi in Stamford is more pronounced than is the case for Milford and Hammonasset. In fact, BSi is slightly lower in Stamford surface waters than in Milford or Hammonasset surface waters; this difference suggests that diatoms in Stamford surface waters are at least as likely to be Si-limited as diatoms in surface waters of Milford and Hammonasset. Furthermore, a larger

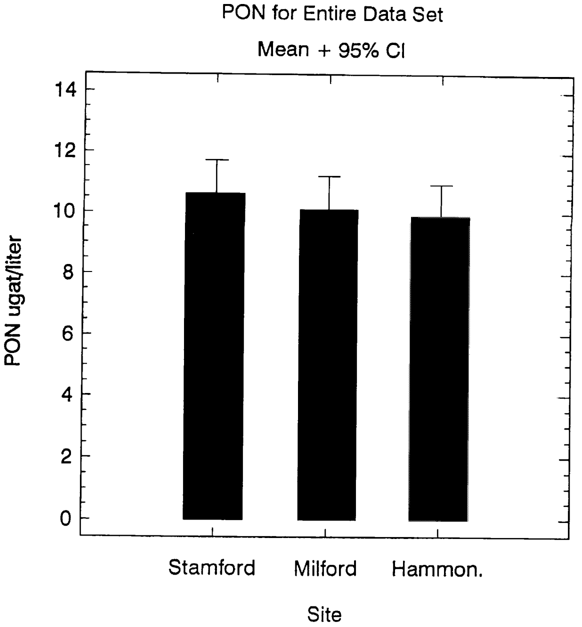

Figure 16. Water Coumn particulate organic nitrogen (PON) averages (μg-atm l^{-1}), over that part of the study when all 3 stations were measured.

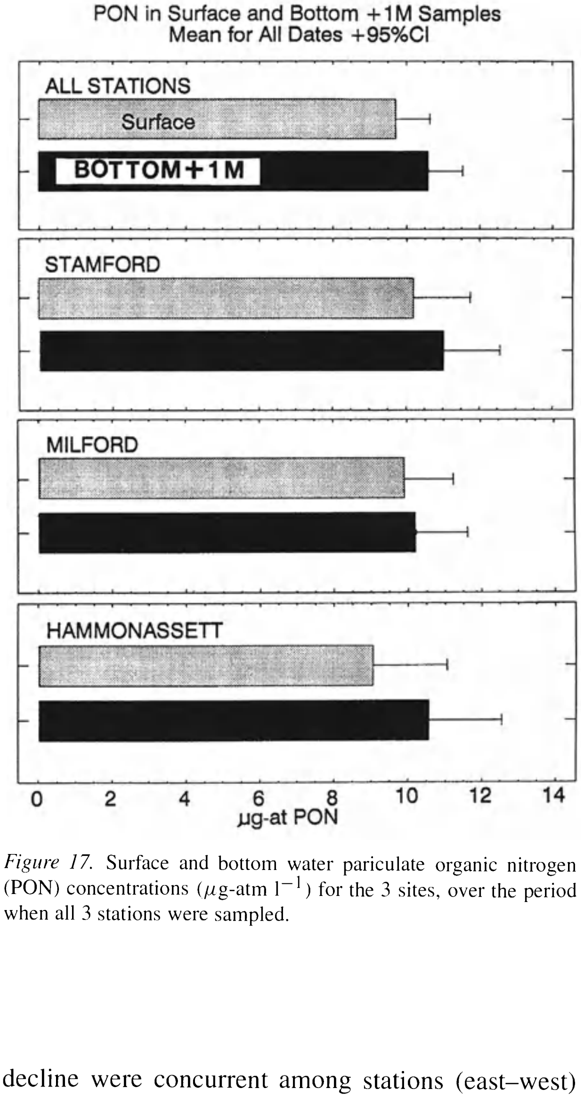

Figure 17. Surface and bottom water pariculate organic nitrogen (PON) concentrations (μg-atm l^{-1}) for the 3 sites, over the period when all 3 stations were sampled.

reservoir of BSi exists in Stamford bottom waters than at the other sites. Assuming that BSi in bottom waters is mainly the result of remineralization of old diatom shells, there is some evidence for enhanced sedimentation of diatoms in western LIS.

Different diatoms, however, have different BSi requirements, often expressed as minimal Si cell quotas, Q_0Si (van Donk & Kilham, 1990). Obviously, larger diatom cells may require more Si per cell than smaller cells, but optimal ratios of Q_0Si: Q_0N for diatoms have been shown to be independent of cell size and species-specific (Tilman et al., 1992), including diatoms isolated from Long Island Sound (Wikfors et al., in review). Questions of Si:N ratios and the importance of lowering the Si:N ratio, by increasing N but not Si, to geographic differences in seasonal phytoplankton community structure in Long Island Sound, are addressed in Wikfors (in review) and Wikfors et al. (in review).

Total & size-fractionated chlorophyll a

Chlorophyll a data

There are four categories of chlorophyll for which data are given: total chlorophyll *a* (Chl), and size fractions of >20 μm (Chl >20), 10–20 μm (Chl 10–20), and <10 μm (Chl <10). All of the more prominent features of strong seasonal cycles of flowering and decline were concurrent among stations (east–west) and between depths (surface–bottom) (Figs 23–26). Minor spatial differences in the cycles did occur, such as comparatively shorter flowerings at Hammonasset, and lack of east–west synchrony of the intermediate chlorophyll fraction (Chl 10–20). Surface and bottom concentrations were well correlated in all categories (<0.01 level of significance), but most strongly in the case of Chl >20. Occurrences of higher chlorophyll in bottom waters (March 1994 for example) could usually be accounted for by Chl >20 (Figs 23–26; Table 8). Occurrences of higher chlorophyll in surface waters (August 1994 at Hammonasset for example) could usually be accounted for by Chl 10–20 and (or) Chl <10 (Figs 23–26 and Table 8). Station-to-station differences in chlorophyll concentrations were clearest when concentrations were high, and less distinct when concentrations were low.

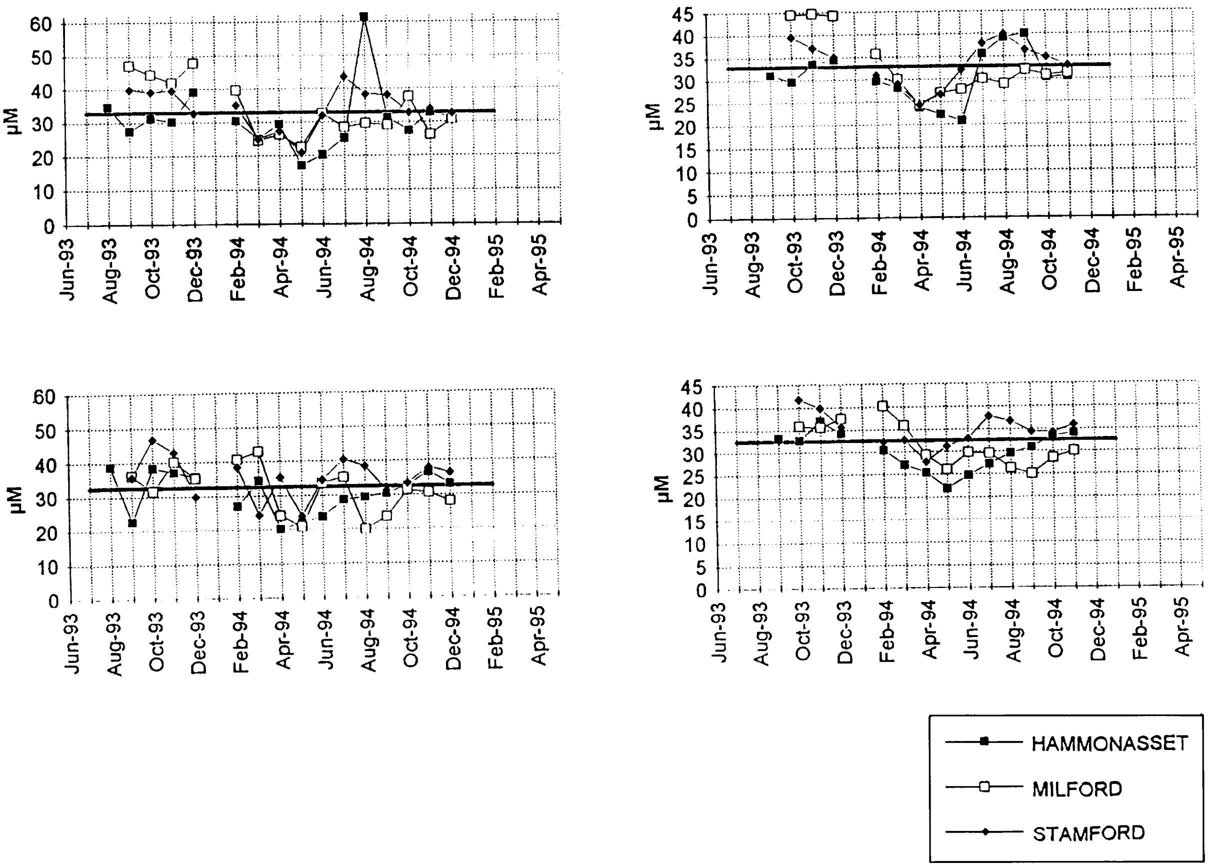

Figure 18. Total nitogen (TN) Concentrations in surface (above) and bottom (below) waters for the 3 study sites. See Figure 10 for data presentation details.

Chlorophyll a

Concentrations ranged between 0.4 and 67 μg l^{-1} at our three stations (Fig. 23). There were peaks in August 1992, April 1993, October 1993, March 1994, August 1994, and April 1995. Peaks in March and April indicate spring blooms, August peaks indicate late summer blooms, and the October peak may be considered a delayed summer, or fall bloom. The August 1992 and April 1993 peaks were lower than the peaks that followed. Spring blooms were less distinct at Hammonasset than at the western stations, as was the October 1993 peak. Hammonasset had the highest concentration in the summer bloom of August 1994. No one station had consistently lowest Chl, but station-to-station differences were consistent enough to produce trends in mean concentration for the period of study (Table 9). There was an increase in the depth average of about 37% between Hammonasset and Milford, and a further increase of about 10% between Milford and Stamford.

Chlorophyll >20μm

Concentrations ranged from 0.2 to 60 μg l^{-1} (Fig. 24). The bulk of the chlorophyll was composed of Chl >20 during winter–spring and fall flowerings, and Chl >20 comprised about 57% of Chl on average at Hammonasset, and about 70% at each of the stations further west, producing the general similarity seen among stations in Figure 23. Seasonal peaks occurred in September 1992, April 1993, October 1993, March–April 1994, October 1994, and March–April 1995. Hammonasset was lowest in the October 1993 peak, the April 1994 peak, the August 1994 peak, and in March but not April of 1995. However, station-to-station differences were consistent enough to produce considerable trends in mean concentration over the period of study (Table 9). There was an overall increase in the depth average of about 61% from Hammonasset to Milford, and an additional small increase of about 5% from Milford to Stamford.

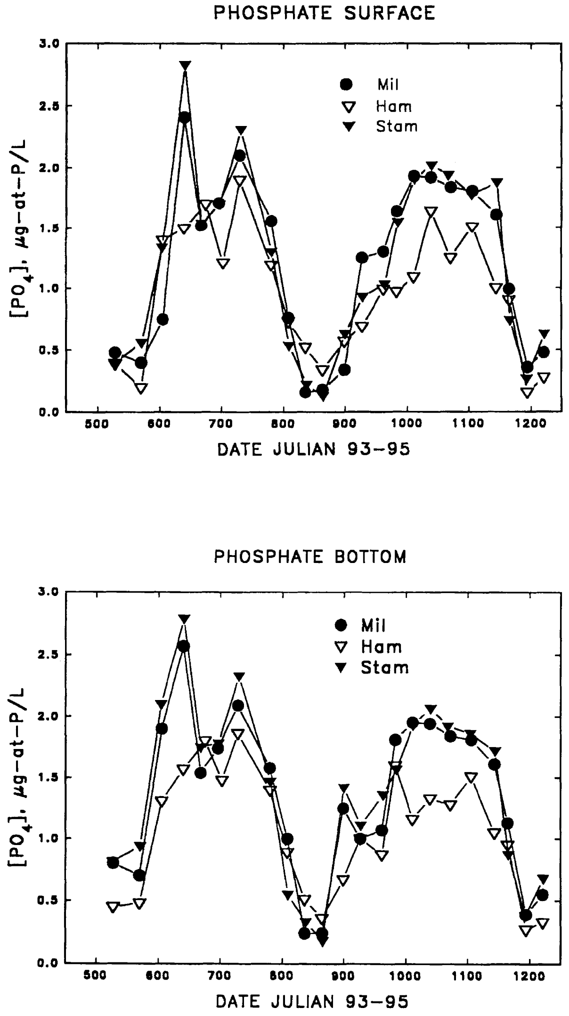

Figure 19. Reactive phosphate concentrations in surface and bottom waters of the 3 study sites.

Chlorophyll 10–20 μm

Concentrations ranged from undetectable to 20 μg l^{-1} (Fig. 26), but only once exceeded 10 μg l^{-1} (August 1994 at Hammonasset). Peaks occurred in August in 1992, 1993, and 1994, and below average values typically occurred from November until March. Although Chl 10–20 comprised on average less than 25% of Chl at Hammonasset, and less than 15% at Milford and Stamford, it made up about half of Chl during the August blooms. No one station had consistently lowest Chl 10-20, however, station-to-station differences were consistent enough to produce trends in mean concentration over the period of study (Table 9). On average, Milford, the center station, had less than Hammonasset, while Stamford was similar to Hammonasset at the surface, and less at the bottom. This reversal of the east–west pattern noted for Chl and Chl >20 is eliminated if the extraordinary peak in Chl 10–20 at Hammonasset in August 1994 is ignored.

Chlorophyll <10 μm

Values ranged from <0.1 to 15 μg l^{-1}, but seldom exceeded 4 μg l^{-1} (Fig. 25). Peaks in Chl <10 occurred around July and August in 1992 and 1994, but not in 1993. This fraction was only about 15 – 20% of Chl on average (Table 8) however, it was as much as 50-75% of Chl in the low months of June and July. The flowering of August 1994 was about 33% Chl <10, the highest percentage observed in any bloom. No one station had consistently lowest Chl <10, however, station-to-station differences were consistent enough to produce overall trends in mean concentration over the period of the study (Table 9). A steady east–west increase occurred at the surface and the bottom.

Chlorophyll summary

Strong seasonal cycles of flowering and decline in chlorophyll stocks occurred concurrently at each station in a manner consistent with the patterns of chlorophyll in temperate estuaries (Riley, 1955). Dominance went to Chl >20 in winter, spring, and fall, and shifted towards the smaller size fractions only in summer. Furthermore, Chl >20 was more abundant on average than the two smaller fractions combined, and it almost entirely dominated all but the August flowerings. There were east–west increases in Chl, Chl >20, and Chl <10 despite more copepod grazers in the west; however, Chl 10–20 did not conform to this pattern. The most conspicuous pattern among stations was the increase between Hammonasset and Milford in the Chl >20 size fraction which dominated Chl, the most positive indication of a eutrophication gradient from east to west in LIS.

Phytoplankton densities

Phytoplankton cell numbers ranged from less than 1 to over 13 million cells per liter in the 1994 Stamford and Hammonasset surface samples counted (Figs 27 and 28). Photosynthetic cells in the <2 μm size range generally represented about 1/3 of the total cell number, as estimated from fluorescence observations; inclusion of the smallest photosynthetic cells in plots did not change seasonal patterns appreciably. The most noteworthy difference in phytoplankton cell numbers between the west and east is not in total counts, but rather in seasonal timing of peaks. Algal cell numbers

Table 4. Mean value and average percentage of total nitrogen for seven nitrogen parameters for the period when all 3 stations were sampled (June 1993 – May 1995) as well as their east–west trends. Row labels are grouped according to the way the parameter was measured as follows: Dir=direct measure; Sum=sum of 2 measures; dif=difference between two measures; Bot=bottom and surf=surface

Parameter measured by			Hammonasset		Milford		Stanford		Trends between stations	
Sum	Dir.	Dif.	Mean µM	%TN	Mean µM	%TN	Mean µM	%TN	Ham-Mil	Mil-Stam
	SURF-TDN		19.9	65	22.4	66	22.7	66	+13%	+1%
	BOT-TDN		18.3	63	19.6	64	22.5	64	+7%	+15%
	SURF-PN		10.2	35	10.7	34	11.4	37	+5%	+7%
	BOT-PN		11.6	37	11.4	36	12.5	36	-2%	+10%
SURF-TN			30.9	100	33.8	100	34.1	100	+9%	+1%
BOT-TN			30.9	100	31.6	100	35.3	100	+3%	+12%
	SURF-NOx		5.8	19	5.6	18	6.3	18	-3%	+13%
	BOT-NOx		5.4	18	6.3	20	6.7	19	+17%	+6%
	SURF-NH3		1.3	4	1.3	4	1.3	4	0	0
	BOT-NH3		1.7	6	1.4	4	1.9	5	–18%	+35%
SURF-DIN			7.1	23	6.9	20	7.6	22	-3%	+10%
BOT-DIN			7.0	23	7.1	22	8.6	24	+1%	+21%
		SURF-DON	11.7	38	14.0	41	13.5	40	+20%	–4%
		BOT-DON	10.4	35	11.4	36	13.2	37	+10%	+16%

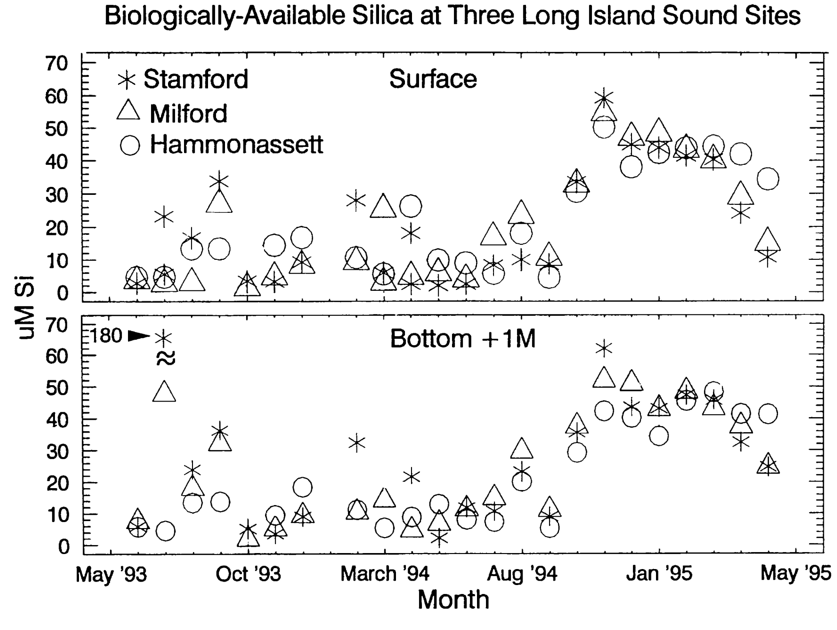

Figure 20. Biologically available Silica (BSi) concentrations in surface and bottom waters of the 3 study sites. Units = µmoles of Silica.

increased in July and August in Hammonasset, and in October in Stamford (Figs 27 and 28).

Considering only the >2 µm phytoplankton, that could be counted in fixed samples, differences in timing of shifts from diatom to non-diatom taxa accompanied seasonal changes in total counts. In Stamford, dinoflagellates replaced diatoms almost entirely in July; at this time, cryptophycean algae bloomed to high numbers in Hammonasset. The accumulation of phytoplankton cells observed in Hammonasset during August and September were mainly dinoflagellates. Differences in peak timing and taxonomic composi-

Table 5. Dissolved inorganic nitrogen to phosphate ratios, and total dissolved nitrogen (TDN) to phosphate ratios for surface and bottom water samples from the 3 stations

Date	Stamford		Milford		Hammonasset	
	Surf	Bot	Surf	Bot	Surf	Bot
N/P Ratios						
6/93	3.3	7.3	4.2	7.1	7.6	9.3
7/93	0.3	5.5	0.8	0.8	9.8	1.3
8/93	1.5	6.6	2.8	2.5	1.3	4.2
9/93	9.5	8.2	6.8	6.8	10	8.7
10/93	4.1	3.6	2.4	2.6	6.0	6.1
11/93	4.0	4.1	3.2	3.4	6.8	5.0
12/93	6.8	7.1	6.1	5.3	5.4	6.1
2/94	5.9	5.6	5.7	6.0	7.2	5.7
3/94	2.4	4.1	2.2	1.3	4.3	3.8
4/94	22.3	2.7	55.8	42.2	20.5	12.8
5/94	1.3	1.0	5.3	0.7	5.3	6.5
93–94 Average	5.6	5.1	8.7	7.1	7.6	6.3
6/94	0.8	2.6	0.8	0.5	4.8	1.7
7/94	0.4	2.6	0.5	0.5	1.3	1.8
8/94	1.3	3.8	1.7	2.7	4.6	3.3
9/94	4.7	3.7	3.1	2.8	3.9	1.6
10/94	9.4	9.0	7.6	7.2	13.1	15.9
11/94	7.6	7.8	6.5	6.5	8.0	7.9
12/94	8.5	8.9	7.2	7.4	10.9	8.9
1/95	10.2	9.4	9.0	8.2	9.1	9.3
2/95	9.5	9.0	10.1	9.4	12.7	11.6
3/95	7.8	9.6	9.9	9.1	10.8	11.0
4/95	3.9	2.8	2.8	4.4	5.5	5.5
5/95	5.3	1.0	6.4	6.2	2.5	7.5
94–95 Average	5.8	5.7	5.5	5.4	7.3	7.2
TDN/P Ratios						
9/93	11.0	10.4	14.0	9.5	15.5	9.2
10/93	16.9	15.0	17.3	9.1	12.8	15.8
11/93	15.9	16.4	18.1	16.2	21.5	20.1
12/93	11.3	9.9	19.1	12.6	15.8	12.0
2/94	17.6	15.7	17.1	17.6	20.4	14.7
3/94	28.7	28.0	13.0	15.1	13.4	14.3
4/94	63.6	63.0	123.0	60.4	52.9	31.2
5/94	91.5	79.4	82.8	56.3	22.4	33.1
93–94 Average	32.1	27.2	38.1	24.6	21.8	18.8
6/94	32.6	16.6	60.0	16.6	24.6	23.7
7/94	26.5	23.1	12.6	21.4	20.7	15.1
8/94	20.5	12.8	7.5	10.0	19.9	12.0
9/94	19.8	12.7	10.5	7.8	22.6	8.5
10/94	14.9	14.9	16.4	11.6	21.1	20.8
11/94	12.9	12.9	10.6	10.9	14.3	14.1
12/94	13.0	15.8	13.4	12.9	18.7	17.9
1/95	14.0	13.4	15.9	12.9	14.4	12.8
2/95	16.5	15.8	17.7	15.6	24.3	21.4
3/95	22.7	20.7	17.8	16.7	22.0	23.1
4/95	36.3	27.9	33.5	30.5	53.8	28.9
5/95	28.0	22.9	40.0	26.7	38.0	27.0
94–95 Average	21.8	17.5	21.3	16.1	24.5	18.8

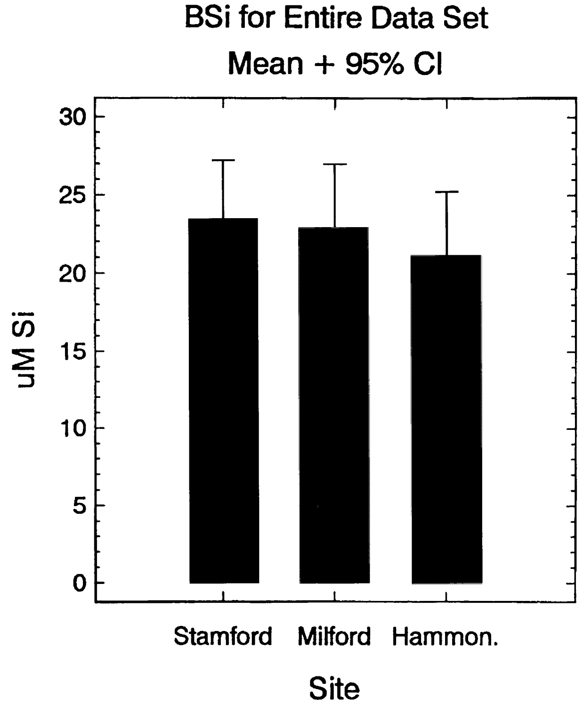

Figure 21. Mean, whole water column, BSi Concentrations averaged over the entire study period for all 3 stations.

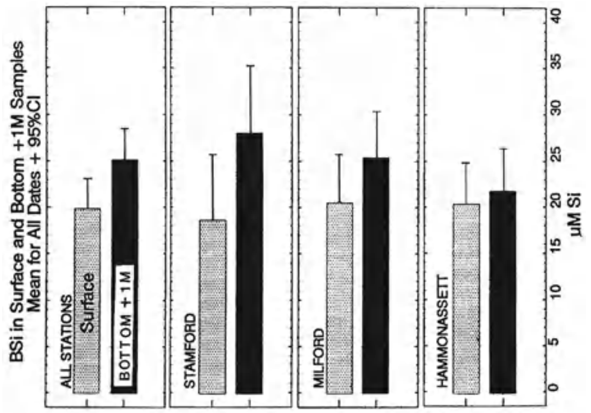

Figure 22. Mean water column biologically available silica concentrations presented separately for surface and bottom water, averaged over the entire study period, for all 3 stations.

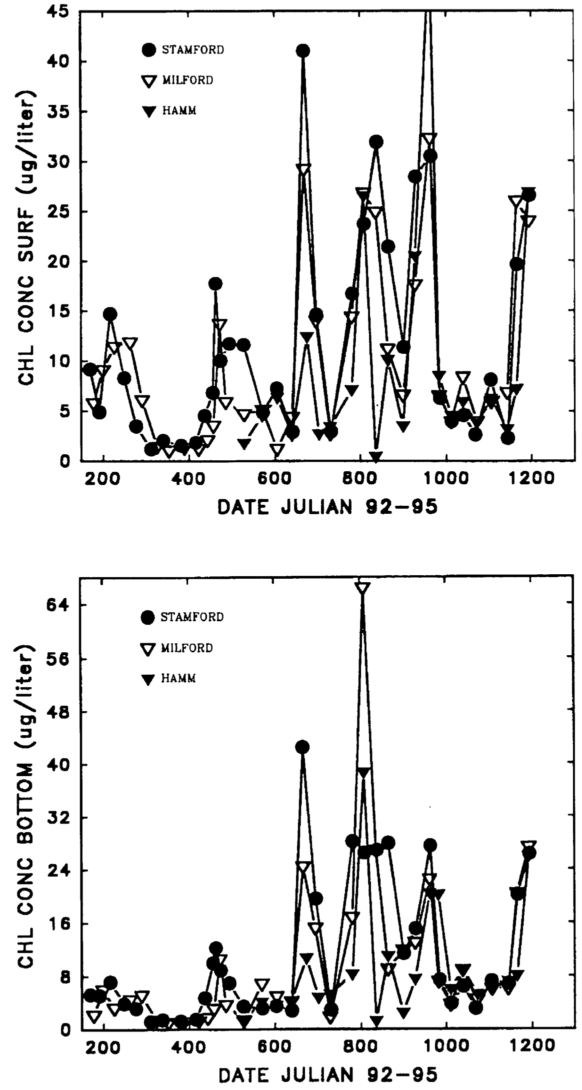

Figure 23. Total surface and bottom water chlorophyll *a* concentrations (as $\mu g\ l^{-1}$) over the entire study, for the 3 stations.

tion of phytoplankton assemblages between Stamford and Hammonasset were clear and pronounced, and are likely controlled by both bottom-up (nutrient) and top-down (grazing) factors.

Photosynthetic nanoplankton (PNANS)

Photosynthetic nanoplankton densities varied from a low of 0.2×10^4 to a high of 7.5×10^4 per ml. (Fig. 29), with well developed seasonal peaks in summer. Densities were higher in surface waters where light is more available. Surface and bottom water seasonal trends were identical as were spatial trends among our 3 sampling sites. Differences among stations were slight but for key summer peaks where the Hammonasset site exhibited concentrations 1.5 – 3 times higher than the more western sites.

Bacterial densities & frequency of dividing cells

Bacterial densities varied from winter lows of about 0.3×10^6 per ml to typical summer highs of $3 - 5 \times 10^6$ per ml in both surface and bottom waters at all 3 of our stations (Fig. 30). Of great interest was the dis-

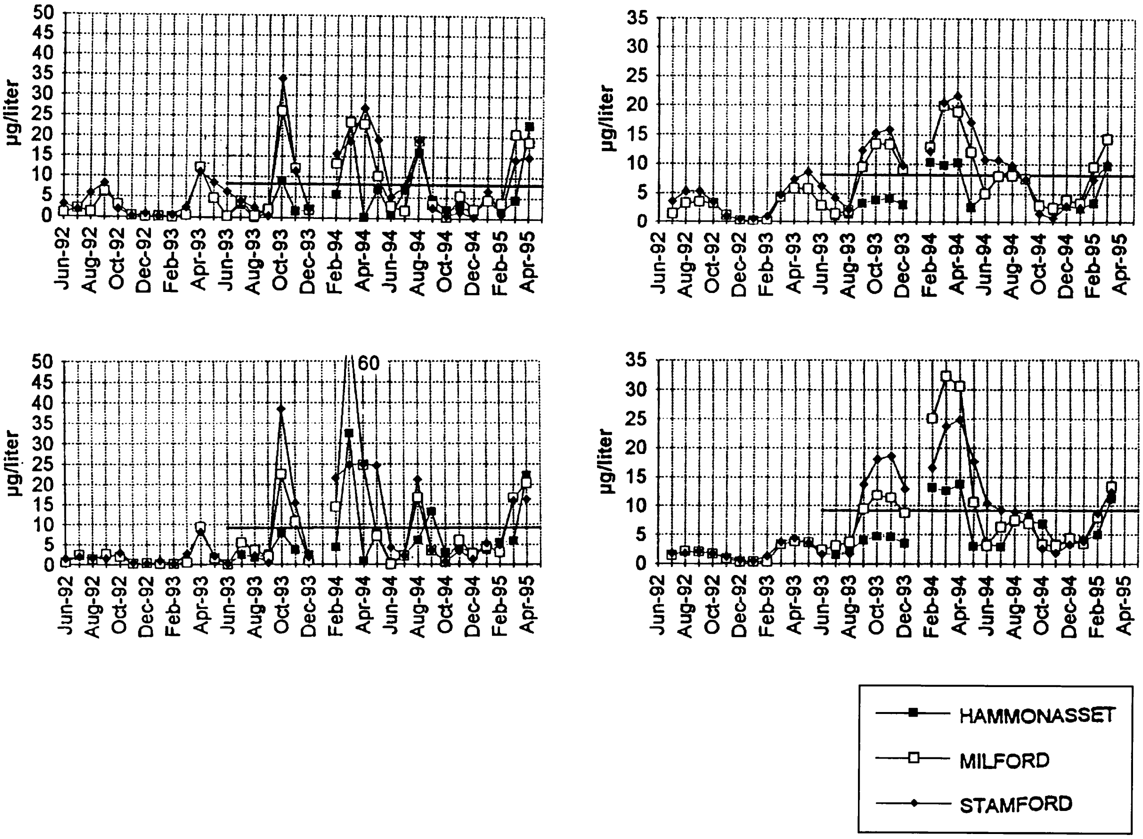

Figure 24. Greater than 20 μm chlorophyll *a* concentrations in surface (above) and bottom (below) waters. See Figure 10 for details on data presentation.

covery of a clear, multi year, repetative seasonal cycle of high *versus* low densities in the surface and bottom waters of all 3 stations. It has been generally assumed until now that no patterns of bacterial abundance exist in coastal marine/estuarine waters. Gradients in abundance do exist from west to east with our western station waters generally exhibiting slightly higher bacterial densities than our eastern station waters, in the summer. No such differences are found between our central and western station waters. One extremely high summer value of 9×10^6 per ml (3 times higher than the other 2 stations) was observed at our western station. This high corresponded to an extreme high silicate value (Fig. 20) and we believe it to be an hypoxic-related effect.

Frequency of dividing cells data, which represent an approximate measure of bacterial growth rates, were similar in surface and bottom waters, and varied between 2% and 7.5% with summer values, in particular, higher in western station waters as compared to eastern station waters (Fig. 31).

Heterotrophic nanoplankton densities

Heterotophic nanoplankton densities were similar in surface and bottom waters of all 3 stations and stations were similar to each other in terms of seasonal patterns. Overall densities on average varied between 0.1×10^3 and 6.4×10^3 per ml, with significant east to west variations at times, and with the central to western station waters sporting higher concentrations than eastern station waters (Fig. 32). Of most note was the extreme interannual differences that appeared between the 1992–93 and 1993–94 sampling seasons.

Phytoplankton species composition & semi-quantitative population estimates

Lists of phytoplankton taxa identified at the three LIS sites during 1993-1995, are presented in Table 10. Total numbers of species found were not remarkably different for different sites; however, identities of dominant taxa varied considerably from site to site. For 1993, during which we sampled Hammonasset

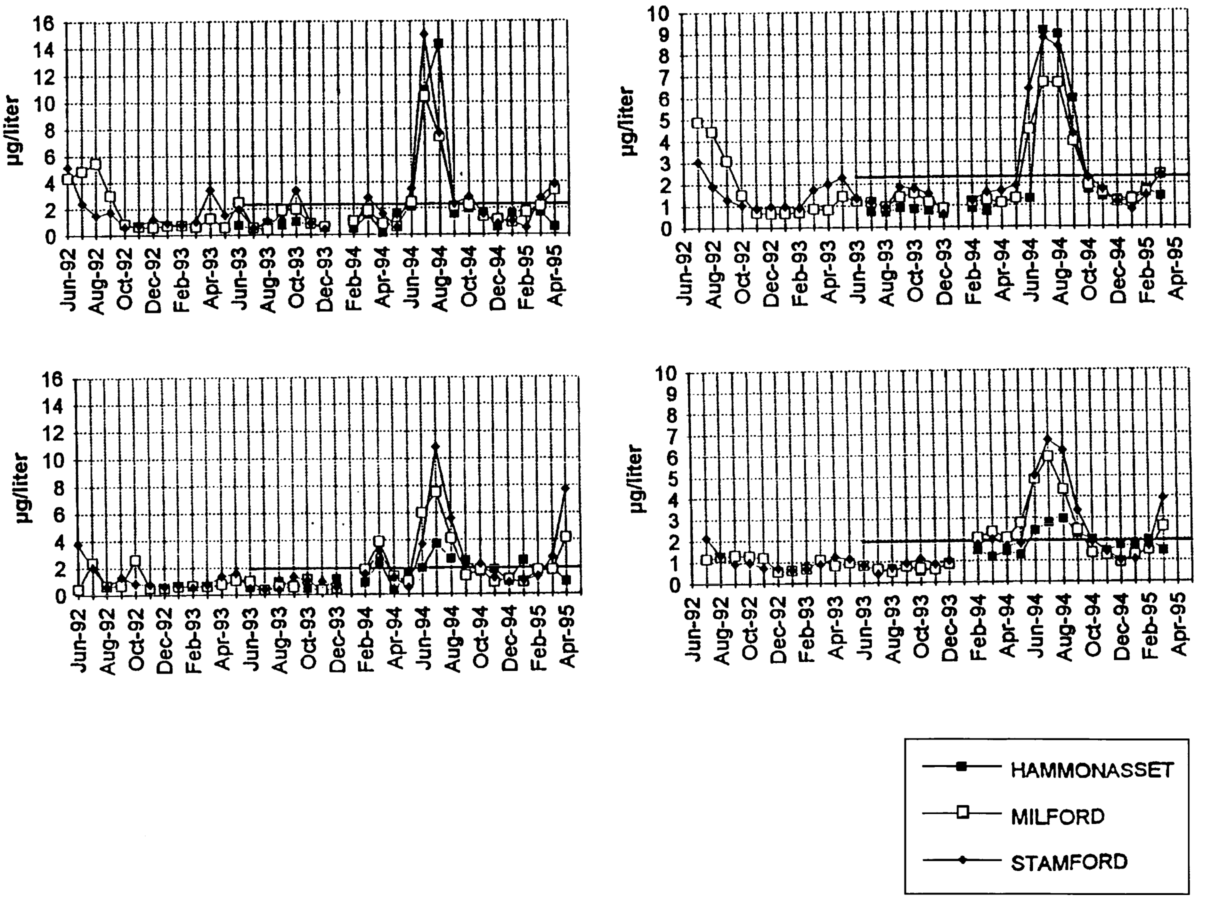

Figure 25. Less than 10 μm chlorophyll *a* concentrations in surface (above) and bottom waters. See Figure 10 for details on data presentation.

for only half the year, eight of 15 dominant phytoplankton taxa in Stamford were not included in the listing of dominants for Hammonasset. Conversely, 7 of 15 Hammonasset dominant taxa were not listed as dominants in Stamford (Table 10). For 1994, the only year with a full sampling schedule, five of 14 Stamford dominants were not even recorded at Hammonasset, whereas, six of 20 Hammonasset dominants were not found in Stamford (Table 10). Finally, in 1995, for which we sampled only winter and spring, nine of 15 Stamford dominants were not dominants in Hammonasset, and two of eight Hammonasset dominants were not dominant in Stamford (Table 10). From this simple listing, it is clear that phytoplankton community structure is different at the three sites.

Percentages of phytoplankton assemblages accounted for by major taxonomic groups also revealed seasonal differences between sites (Fig. 33). For all three sites, centric diatoms tend to dominate the winter and early spring flora, but pennate diatoms occasionally contribute considerably, especially in bottom waters where benthic taxa may be mixed into the water column. Dinoflagellates showed a major pulse in Stamford and Milford during the summer of 1992, with more modest increases in 1993–1994. Hammonasset experienced a significant increase in dinoflagellates during the summer of 1994, and overall had study-averaged lower concentrations of dinoflagellates than either the central or western stations (Fig. 33). Green algae and crytophytes remained only minor floral constituents, with one or two exceptions.

Occasionally, e.g. spring of 1993 at Stamford and Milford and August of 1994 in Hammonasset, picoplanktonic cells (<2 μm) showed strong dominance; it is impossible to identify these cells with light or fluorescent microscopy. In most cases, bottom and surface waters tracked each other fairly well, resulting from lack of stratification and deposition of surface algae to bottom waters. An exception to this is that dinoflagellates tended to be slightly more abundant in

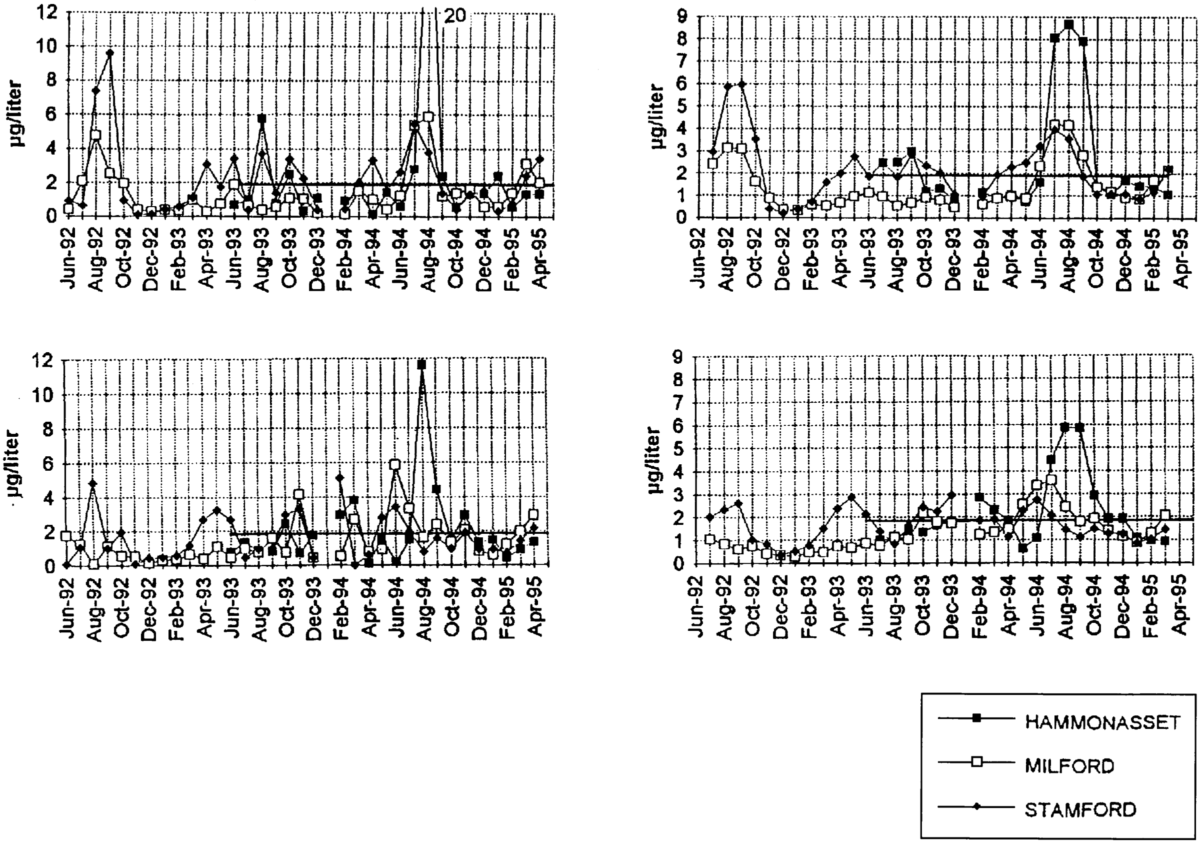

Figure 26. Ten –20 μm chlorophyll *a* concentrations in surface (above) and bottom (below) waters. See Figure 10 for details on data presentation.

surface waters than in bottom, resulting possibly from depth regulation by the dinoflagellates themselves or lower sinking rates because of less grazing. Seasonal shifts in major phytoplankton groups generally followed expected patterns, but differences in identities of dominant taxa and timing of shifts were apparent for the different sites.

Water column ciliate species composition

Prior to this study about 28 species of water column ciliates (mostly tintinnids) have been reported for the waters of Long Island Sound (Capriulo & Carpenter, 1983). This study extends the list of species to 71 (Table 11). Differences in species composition among stations were found on most of the sampling dates. A summary of the 1993–94 and the 1994–95 sampling periods indicates that of the 71 species encountered 94% of them were observed in our western station waters, while 80% occurred in our central station waters and only 62% in the waters of our eastern most station (Table 12). Additionally, some differences were encountered between surface and bottom waters of the same station for all 3 stations. Seasonal data on when various species of ciliates were present in the Sound at each of the respective sites will be presented in a future contribution (Capriulo & Pellet, in prep.).

Copepod abundance & species composition

Standing stocks varied between 183 and 64 152 individuals per cubic meter (Table 13). There were pronounced annual cycles in copepod abundance (Fig. 34). Stocks were generally high from mid-spring through early-summer, and generally low from mid-summer through winter. The amplitude of the seasonal cycle increased west of Hammonasset, and the range at the western station was about five times that of Hammonasset. The arithmetic and geometric means (Table 13 and Fig. 34) suggested a sharp increase in copepods between Hammonasset and Milford, but not much difference between Milford and Stamford.

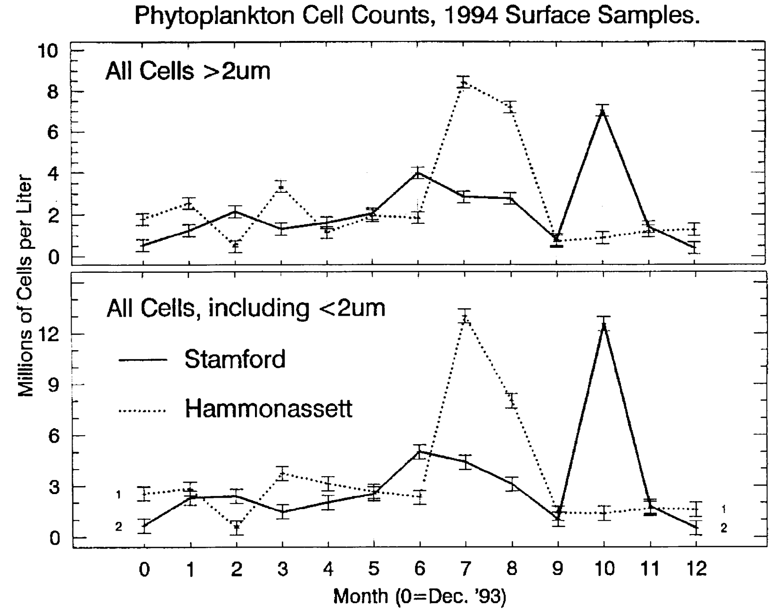

Figure 27. Quantitative phytoplankton cell counts for 1994 surface samples from the Stamford and Hammonasset stations, for all cells greater than 2 μm (top), and all cells including the less than 2 μm (bottom) sizes.

Table 6. Whole water column dissolved inorganic nitrogen to phosphate ratios, and total dissolved nitrogen (TDN) to phosphate ratios, for the 3 study sites

	Stamford	Milford	Hammonasset
Dissolved inorganic nitrogen/phosphate ratio			
1993–1994			
Average	5.4	7.9	7.0
1994-1995			
Average	5.8	5.5	7.3
1993–1995			
Average	5.4	6.3	7.2
Total dissolved nitrogen/phosphate ratio			
1993–1994			
Average	29.7	31.4	21.1
Median	16.7	17.2	17.8
1994-1995			
Average	19.7	18.7	21.7
Median	16.5	15.7	21.1
1993–1995			
Average	24.1	23.8	21.1
Median	16.5	16.3	20.3

Table 7. Correlations between surface water and bottom water concentrations for total chlorophyll as well as each of the 3 size fractions of chlorophyll examined in this study

Parameter	*r*	*p*
Chl	0.80	<0.01
Chl >20	0.88	<0.01
Chl 10–20	0.63	<0.01
Chl <10	0.75	<0.01

The amount of increase between Hammonasset and Milford appeared to be affected by season, since the difference was most noticeable in the late spring. The highest numbers in all statistical categories (Table 13) occurred at Milford rather than Stamford.

The possibility of net clogging was evaluated during 37 of the 102 tows. The ratio of a flow meter positioned inside the mouth of the net to a meter positioned outside provided rough estimates of the nets filtration efficiency (UNESCO, 1968). Those ratios varied from a minimum of 0.38 to a maximum of 1.06, with a mean of 0.83 and a median of 0.84. It is arguable that filtration efficiencies estimated by inside and outside flowmeters are biased towards lower values. The nets forward motion can displace and accelerate water, causing an increase in the rate of flow through the outside flowmeter as the flow through the inside decreases (UNESCO, 1968). Nevertheless, the

Table 8. Chlorophyll size-fractionation data (for the 3 size-fractions: >20, 10–20, and <20 μm) as percentage of total

Month	Size-Fractionated Chlorophyll Percentages								
	Stamford			Milford			Hammonasset		
	>20 μm	10–20 μm	<10 μm	>20 μm	10–20 μm	<10 μm	>20 μm	10–20 μm	<10 μm
Surface									
6/18/92	34.0	10.0	56.0						
6/26/92				18.0	7.0	75.0			
7/9/92	38	13.0	49.0						
7/17/92				23.0	23.0	54.0			
8/3/92	39.5	5.0	10.5						
8/12/92				10.6	42.0	47.6			
9/4/92									
9/18/92				53.0	22.0	25.0			
10/2/92	55.0	27.0	18.0						
10/16/92				48.0	32.0	14.0			
11/6/92	29.0	8.0	63.0						
11/20/92				29.0	26.0	45.0			
12/4/92	45.0	5.0	60.0						
12/18/92				22.0	25.0	53.0			
1/15/93	14.0	27.0	59.0						
1/23/93				16.0	29.0	55.0			
2/19/93	27.0	31.0	42.0						
2/26/93				11.0	30.0	59.0			
3/12/93	53.0	25.0	22.0						
3/19/93				25.0	46.0	29.0			
3/31/93	72.0	7.0	21.0						
4/2/93				71.0	13.0	16.0			
4/7/93	63.0	18.0	19.0						
4/18/93	35.0	25.0	40.0						
4/30/93				77.0	13.0	10.0			
5/8/93	72.0	15.0	13.0						
6/10/93	54.0	30.0	17.0						
6/11/93				7.0	40.0	53.0	18.0	38.0	43.0
7/23/93				76.0	13.0	11.0	67.0	22.0	11.0
7/24/93	81.0	8.0	11.0						
8/26/93	34.0	51.0	15.0						
8/27/93				36.0	32.0	32.0	0	86.0	14.0
10/1/93				45.0	13.0	43.0	45.0	28.0	27.0
10/2/93	13.0	48.0	39.0						
10/29/93	84.0	8.0	8.0	90.0	4.0	7.0			
11/5/93							72.0	20.0	8.0
11/26/93				86.0	7.0	6.0			
11/27/93	78.0	16.0	6.0						
12/3/93							61.0	12.0	27.0
12/29/93				70.0	7.0	23.0	52.0	32.0	16.0
12/31/93	72.0	13.0	15.0						
2/18/94				92.0	1.0	7.0	80.0	13.0	7.0
2/19/94	95.0	3.0	3.0						
3/18/94				88.0	5.0	7.0	88.0	6.0	6.0
3/19/94	80.0	8.0	12.0						

Continued on p. 303

Table 8. contd.

Month	Size-Fractionated Chlorophyll Percentages								
	Stamford			Milford			Hammonasset		
	>20 μm	10–20 μm	<10 μm	>20 μm	10–20 μm	<10 μm	>20 μm	10–20 μm	<10 μm
4/15/94				92.5	4.1	3.4	41.6	30.5	27.8
4/17/94	88.0	7.0	5.0						
5/13/94				91.0	3.6	5.5	69.5	14.2	16.3
5/14/94	90.2	7.3	2.7						
6/17/94				43.4	24.1	32.5	22.1	15.2	62.6
6/18/94	45.6	23.3	31.1						
7/15/94				10.3	30.7	59.0	30.3	14.1	55.6
7/16/94	28.0	19.2	52.8						
8/19/94				61.5	14.8	23.7	31.5	54.5	13.9
8/21/94	62.3	12.5	25.2						
9/9/94				49.3	17.9	32.8	53.6	28.3	18.1
9/11/94	40.5	22.3	37.2						
10/7/94				6.9	35.5	57.6	45.5	11.9	42.6
10/8/94	17.3	10.8	71.9						
11/4/94				64.6	18.6	16.8	47.5	24.2	28.3
11/5/94	36.1	29.5	34.4						
12/3/94	28.2	39.2	32.6						
12/7/94				56.1	15.2	28.8	48.8	36.0	15.2
1/9/95	83.6	4.1	12.3						
1/10/95				74.6	9.9	15.5	68.3	12.9	18.8
2/17/95				55.6	20.1	24.3	55.6	21.5	22.9
2/18/95	45.5	32.7	21.9						
3/10/95				79.5	12.1	8.3	62.9	22.0	15.1
3/11/95	73.4	12.3	14.3						
4/7/95				77.9	8.0	14.0	85.4	7.0	7.6
4/8/95	56.6	13.0	30.4						
5/5/95				10.8	42.6	46.7	64.8	16.3	18.9
5/6/95	21.4	6.8	71.8						
Bottom									
6/18/92	30.0	1.9	71.9						
7/9/92	40.0	21.0	39.0						
7/17/92				39.0	21.0	40.0			
8/3/92	23.0	68.0	9.0						
9/4/92	40.0	26.0	34.0						
9/18/92				59.0	25.5	15.5			
10/2/92	39.9	8.4	52.0						
10/16/92				39.0	11.0	50.0			
11/6/92	17.4	31.0	51.4						
11/20/92				24.0	39.6	36.7			
12/4/92	9.0	41.0	50.0						
12/18/92				37.5	13.5	49.0			
1/15/93	50.0	25.0	16.0						
1/23/93				16.0	30.0	54.0			
2/19/93	68.0	17.0	15.0						
2/26/93				17.0	30.0	53.0			
3/12/93	67.0	22.0	11.0						
3/19/93				28.0	38.0	34.0			
3/31/93	89.0	4.0	7.0						
4/7/93	61.0	11.0	28.0						
4/16/93				74.0	7.0	19.0			

Continued on p. 304

Table 8. contd.

Month	Size-Fractionated Chlorophyll Percentages								
	Stamford			Milford			Hammonasset		
	>20 μm	10–20 μm	<10 μm	>20 μm	10–20 μm	<10 μm	>20 μm	10–20 μm	<10 μm
4/18/93	31.0	46.0	23.0						
4/30/93				42.0	30.0	28.0			
6/10/93	9.0	77.0	14.0						
6/11/93				0	33.0	67.0	12.0	53.0	35.0
7/23/93				80.0	15.0	5.0	61.0	33.0	6.0
7/24/93	72.0	16.0	13.0						
8/26/93	63.0	27.0	10.0						
8/27/93				73.0	15.0	12.0	49.0	25.0	26.0
10/1/93				50.0	38.0	13.0	63.0	20.0	17.0
10/2/93	18.0	36.0	47.0						
10/29/93	91.0	7.0	2.0	92.0	3.0	5.0			
11/5/93							73.0	23.0	4.0
11/26/93				70.0	27.0	2.0			
11/27/93	78.0	17.0	5.0						
12/3/93							76.0	15.0	9.0
12/29/93				52.0	25.0	23.0	46.0	33.0	21.0
12/31/93	67.0	15.0	18.0						
2/18/94				86.0	3.0	11.0	53.0	36.0	11.0
2/19/94	77.0	18.0	5.0						
3/18/94				90.0	4.0	6.0	84.0	10.0	6.0
3/19/94	88.0	0	12.0						
4/15/94				92.0	3.0	5.0	67.3	10.3	22.4
4/17/94	93.0	2.3	4.6						
5/13/94				78.8	10.4	10.7	71.4	13.9	14.7
5/14/94	88.2	10.0	2.0						
6/17/94				0	49.0	51.0	14.1	8.8	77.1
6/18/94	37.5	30.0	32.5						
7/15/94				16.9	25.4	57.7	30.9	20.1	49.0
7/16/94	15.2	13.1	71.7						
8/19/94				74.4	7.2	18.4	29.9	57.1	13.0
8/21/94	76.9	2.5	20.6						
9/9/94				47.8	33.2	19.0	65.6	20.4	14.0
9/11/94	48.4	21.4	30.2						
10/7/94				16.1	35.8	48.1	49.0	23.9	27.1
10/8/94	18.6	30.6	50.8						
11/4/94				66.4	23.5	10.1	47.3	32.7	20.0
11/5/94	51.8	29.2	19.0						
12/3/94	43.5	30.1	26.4						
12/7/94				58.0	18.3	23.8	56.8	25.9	17.2
1/9/95	73.7	12.8	13.5						
1/10/95				75.6	9.7	14.7	61.2	24.4	14.4
2/17/95				50.9	20.2	28.9	76.5	6.0	17.6
2/18/95	70.6	10.3	19.0						
3/10/95				81.5	9.8	8.8	73.6	11.9	14.5
3/11/95	79.1	7.2	13.6						
4/7/95				74.1	10.7	15.3	5.3	85.7	9.0
4/8/95	61.8	8.2	29.3						
5/5/95				25.4	23.6	51.0	63.4	23.5	13.1
5/6/95	19.9	13.2	66.9						

Table 9. Surface and bottom water mean chlorophyll *a* concentrations and the percentage contribution of each size-fraction to the total chlorophyll during the time when all 3 stations were sampled

	Hammonasset		Milford		Stamford		Trends Between Stations	
	Mean-μgl^{-1}	%TOT	Mean-μgl^{-1}	%TOT	Mean-μgl^{-1}	%TOT	Ham-Mil	Mil-Stam
SURF Chl	9.9	100	12.8	100	14.5	100	+30%	+14%
BOT Chl	9.5	100	13.7	100	14.2	100	+44%	+4%
SURF Chl >20	5.5	56	8.7	68	9.5	66.0	+58%	+8%
BOT Chl >20	6.1	58	10.0	73	10.3	73.0	+64%	+3%
SURF Chl 10–20	2.3	23	1.6	13	2.0	14.0	−30%	+25%
BOT Chl 20–20	2.0	21	1.7	12	1.6	11.0	−15%	−5%
SURF Chl <10	2.1	21	2.3	17	2.9	20.0	+10%	+26%
BOT Chl <10	1.4	15	2.0	15	2.4	17.0	+43%	+20%

accuracy of the copepod abundance data was likely compromised now and then.

Intrastation patchiness, based on repeated tows done on 13 occasions, ranged from strong to weak. The data were binned, and this treatment indicated that patchiness was most intense when copepod abundance was 1000–5000 individuals per cubic meter, suggesting that differences between stations would be hardest to detect within that range. In any case, the copepods were usually significantly patchy, and since patchiness data was available for only 13 of 93 samples, comparisons of copepod abundance among stations on individual dates are more-or-less uncertain. Comparing mean abundance among the stations eliminates this problem because every month represents a replicate, and the average implicitely incorporates random errors like those due to patchiness and sampling.

Tests of copepod distribution

Two-way ANOVA (Zar, 1984) was used to test the significance of station location and time of year as factors determining copepod abundance. The data were transformed (natural log), and tested acceptably for normality (Kolmogorov-Smirnoff test p=0.45), and homogeneity of variance (Cochran's C test p=0.51, and Bartlett's test p=0.35), thus meeting the underlying assumptions for ANOVA (Zar, 1984). Station location influenced copepod abundance significantly (p=0.0035). *A posteriori* testing (Zar, 1984) indicated that Hammonasset had fewer copepods than Milford and Stamford ($p \leq 0.05$), while Milford and Stamford were not different. In addition, a nonparametric Kruskall–Wallis test (Zar, 1984) ranked Hammonasset lowest in copepod abundance ($p<0.001$). The seasonal cycle was also significant ($p<0.001$). The data were organized in four seasonal bins (Jan.+ Feb.+ Mar.= winter, and so on), and *a posteriori* testing ranked spring highest in copepods, while summer

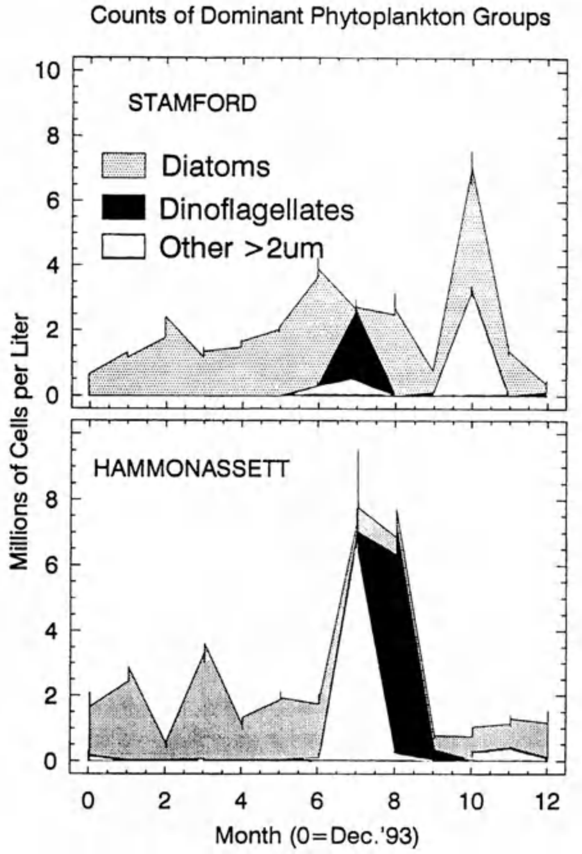

Figure 28. Quantitative counts of dominant phytoplankton by groups (diatoms, dinoflagellates and other greater than 2 μm forms), for the December 1993 to December 1994 sampling period at the Stanford and Hammorasset sites.

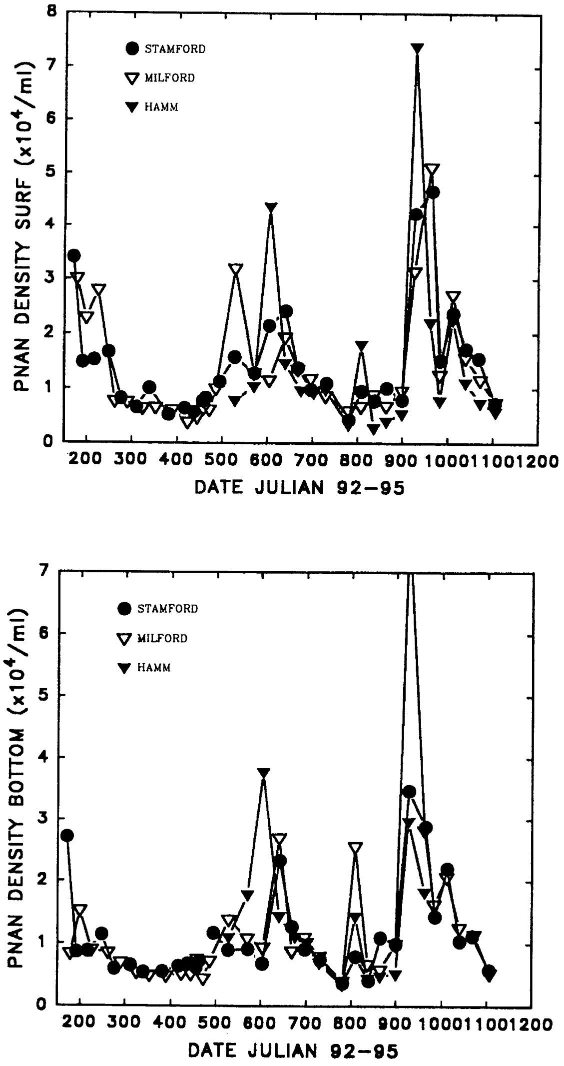

Figure 29. Photosynthetic nanoplankton (PNAN) concentrations for the 3 stations over the entire study period, for surface and bottom waters.

and fall shared lowest rank ($p \leq 0.05$). The interaction between station and season was not significant (p=0.525) even though it appeared that differences among stations were greatest in late spring.

In summary, Hammonasset, the eastern station, had fewest copepods. Spatial gradients seemed less intense during lows in the annual abundance cycle, but interstation comparisons on any given date were suspect because of intrastation patchiness. Nevertheless, when station means were tested, the difference between Hammonasset and the western stations was significant, whereas there was no discernable difference between the two western stations.

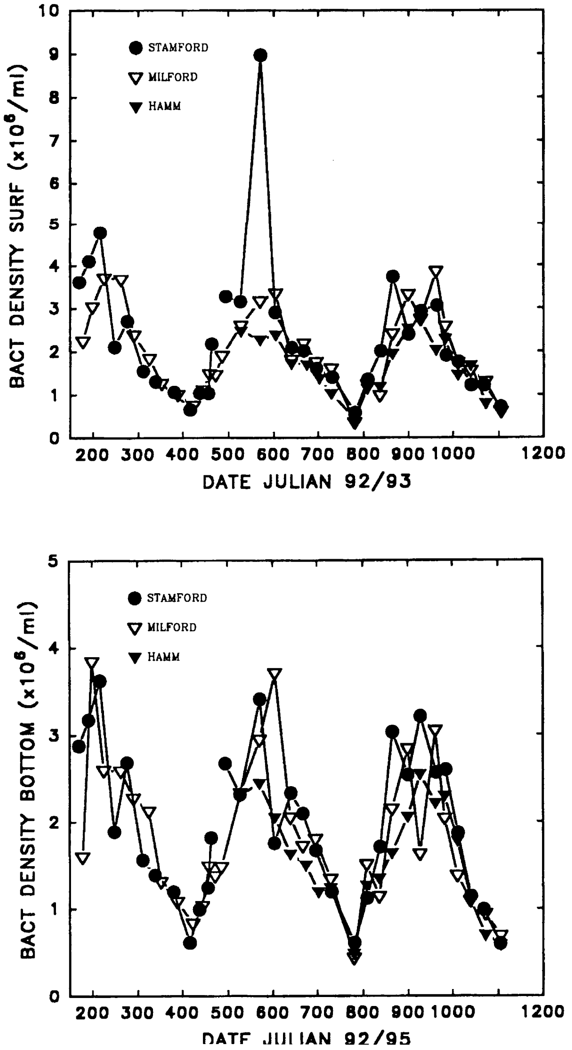

Figure 30. Planktonic bacterial concentrations for the 3 study sites over the entire study period, for surface and bottom waters.

Copepod biomass

Not surprisingly, seasonal cycles and spatial trends in copepod biomass (Figs 35 and 36, Table 14) matched well with cycles and trends in copepod abundance. The data suggest an overall increase between Hammonasset and points to the west, and there were high seasonal peaks every spring, most especially in the west. Seasonal variability in biomass became more extreme from east to west, ranging from two to four orders of magnitude at the different stations (Table 14). During spring peaks, there were more of the heavy copepod, *Temora longicornis*, at the western stations, making east–west differences in biomass greater at times than east–west differences based on abundance.

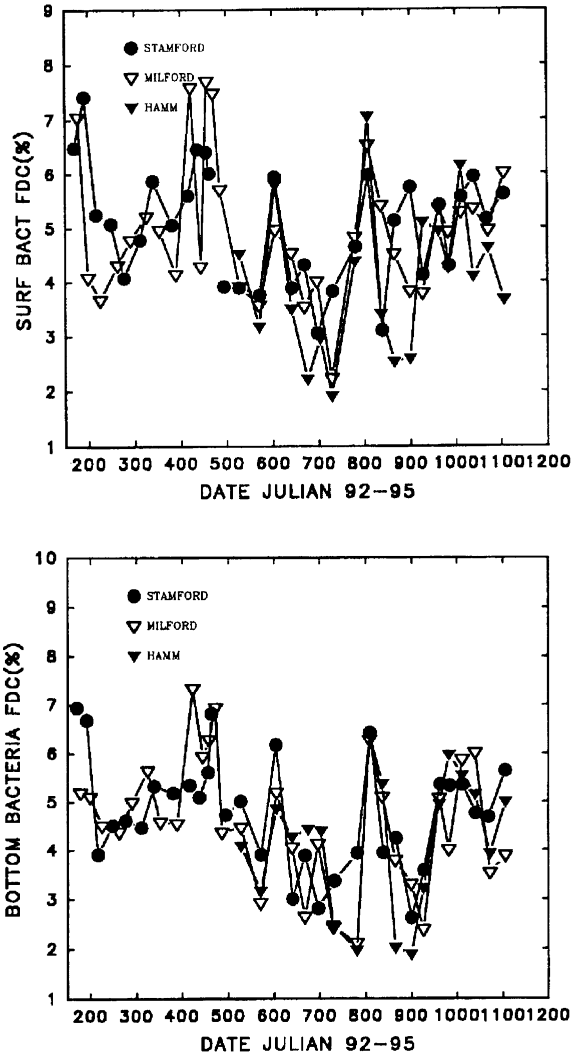

Figure 31. Bacterial growth rates estimated as frequency of dividing cells (FDC), for the 3 sites over the entire study period, for surface and bottom waters.

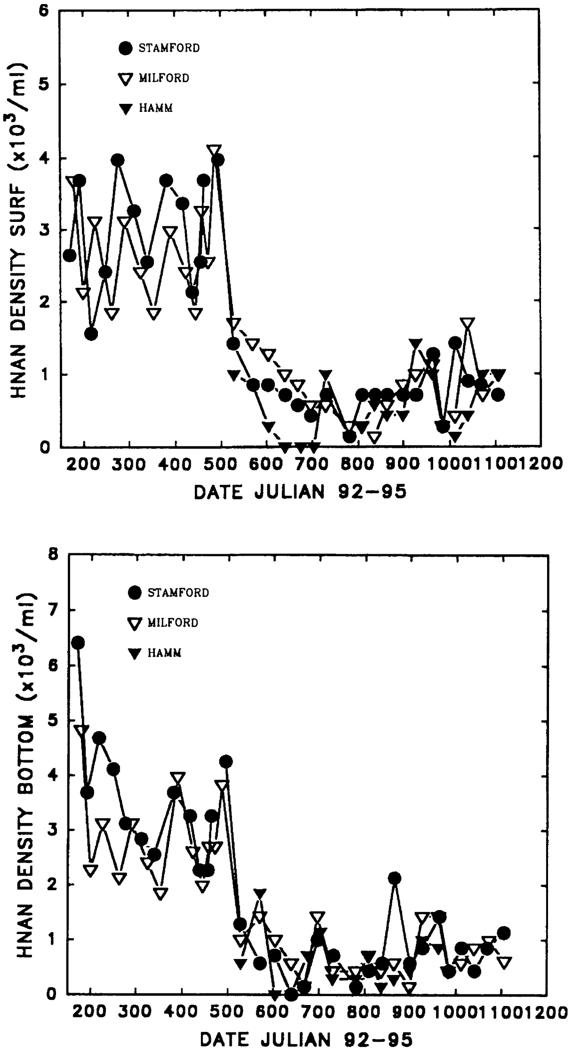

Figure 32. Heterotrophic nanoplankton (HNAN) concentrations for the 3 study sites over the entire study period, for surface and bottom waters.

Conversely, there were instances during seasons of low abundance when Hammonasset had greater biomass, and fewer, heavier, individuals than western stations. In fact, individual *Temora longicornis, Acartia hudsonica,* and *Acartia tonsa* at Hammonasset appeared to weigh 10–20% more on average than they did at Milford and Stamford. This suggests a difference among stations in the age (growth stage) distribution of the copepods within the population.

Tests of biomass distribution

Two-way ANOVA (Zar, 1984) was applied to biomass just as it was to abundance. The data were not normally distributed, so they were transformed (natural log), after which they passed tests for normality (Kolmogorov–Smirnoff test p=0.86), and homogeneity of variances (Cochrans' C test p=0.46, and Bartletts' test p=0.24). Season was a significant influence on biomass (p<0.001), and *a posteriori* testing confirmed that biomass was highest in spring (p<0.05). During the summer, biomass was generally low, and monthly spatial patterns did not suggest consisten increases from east to west. Differences in mean biomass among stations achieved a significance level approaching 5% (p=0.057) only when the summer data were excluded from the test. *A posteriori* testing confirmed that Hammonasset had less biomass than

Table 10. Phytoplankton species list for all 3 stations for the 1993, 1994, and 1995 study period, indicating the relative abundance as + or ++

Class	Genus	Species	Hammonasset*	Milford	Stamford
1993 Phytoplankton Species List					
Dinophyceae	Amphidinium	sp.			+
Bacillariophyceae	Amphiprora	sp.		++	
Bacillariophyceae	Amphora	sp.	+	+	+
Bacillariophyceae	Asterionella	japonica	+	+	
Bacillariophyceae	Biddulphia	sp.		+	+
Dinophyceae	Ceratium	sp.		+	
Bacillariophyceae	Chaetoceros	affinis	+		+
Bacillariophyceae	Chaetoceros	cuvisetus	++	++	+
Bacillariophyceae	Chaetoceros	danicus		+	
Bacillariophyceae	Chaetoceros	decipiens			+
Bacillariophyceae	Chaetoceros	lorenzianus			+
Bacillariophyceae	Chaetoceros	simplex	+	+	+
Bacillariophyceae	Chaetoceros	tortissimus			+
Chlorophyceae	Chlorella	sp.	+		
Prymnesiophyceae	Coccolithus	huxleyii		+	
Bacillariophyceae	Coscinodiscus	sp.	++	++	+
Chlorophyceae	Cryptomonas	sp.	++	++	
Bacillariophyceae	Cyclotella	cryptica	+	++	
Bacillariophyceae	Detonula	confervaceae	+		+
Silicoflagellate	Dictyocha	sp.			+
Dinophyceae	Dinophysis	acuminata	+	+	+
Silicoflagellate	Distephanus	speculum		++	++
Bacillariophyceae	Ditylum	brightwellii	+	+	+
Bacillariophyceae	Euchampia	zoodiacus	++		++
Euglenophyceae	Euglena	sp.	+		
Bacillariophyceae	Grammatophora	sp.	+	+	
Dinophyceae	Gymnodinium	sanguineum	+	+	
Bacillariophyceae	Gyrosigma	sp.	++		
Bacillariophyceae	Hemiaulis	sinensis	+	+	
Dinophyceae	Heterocapsa	triquetra	+		+
Bacillariophyceae	Heterosigma	sp.			+
Bacillariopnyceae	Leptocylindrus	danicus	++	++	++
Bacillariophyceae	Leptocylindrus	minimum	+	++	++
Bacillariophyceae	Licmophora	sp.			+
Bacillariophyceae	Melosira	sulcata	+	++	++
Bacillariophyceae	Navicula	sp.	++	+	++
Bacillariopjyceae	Nitzschia	closterium		+	
Bacillariophyceae	Nitzschia	longissima	+	+	++
Bacillariophyceae	Nitzschia	seriata	++	++	++
Bacillariophyceae	Nitzschia	sp.	++		+
Dinophyceae	Oxytoxum	sp.		+	
Prymnesiophyceae	Pavlova	sp.	+	+	+
Dinophyceae	Peridinium	sp.		+	
Prymnesiophyceae	Phaeocystus	pouchetti		+	
Dinophyceae	Prorocentrum	micans		+	+
Dinophyceae	Prorocentrum	minimum		++	+
Dinophyceae	Prorocentrum	scutellum	+	+	++
Dinophyceae	Prorocentrum	triestinum		+	+
Dinophyceae	Protroperidinium	sp.		+	+
Prymnesiophyceae	Pyramimonas	sp.	+	+	

Continued on p. 309

Table 10. contd.

Class	Genus	Species	Hammonasset*	Milford	Stamford
Bacillariophyceae	Rhizosolenia	delicatula			++
Bacillariophyceae	Rhizosolenia	faeroense		+	++
Bacillariophyceae	Rhizosolenia	setigera		+	+
Bacillariophyceae	Rhizosolenia	stolterfothii			+
Bacillariophyceae	Skeletonema	costatum	++	++	++
Bacillariophyceae	Stephanopyxix	sp.	+	+	
Prymnesiophyceae	Tetraselmis	sp.		+	
Bacillariophyceae	Thalassionema	nitzschioides	++	+	
Bacillariophyceae	Thalassiosira	decipiens		+	
Bacillariophyceae	Thalassiosira	gravida		+	
Bacillariophyceae	Thalassiosira	nordenskoldii	++	++	++
Bacillariophyceae	Thalassiosira	pseudonana	++	++	++
Bacillariophyceae	Thalassiosira	rotula	++	++	+
Bacillariophyceae	Thalassiothrix	frauenfeldii	++	++	++
Bacillariophyceae	Thalassiothix	longissima	+	+	+
Total number of species by station, 1993			36	48	45
1994 Phytoplankton Species List					
Bacillariophyceae	Achnanthes	sp.	+	+	
Dinophyceae	Amphidinium	sp.	+	+	
Bacillariophyceae	Amphiprora	sp.	+		
Bacillariophyceae	Asterionella	japonica	++	++	++
Bacillariophyceae	Chaetoceros	debilis		+	+
Bacillariophyceae	Chaetoceros	simplex	+	+	
Chlorophyceae	Chlorella	sp.		+	
Prymnesiophyceae	Coccolithus	huxleyii		+	
Bacillariophyceae	Coscinodiscus	sp.	++	+	++
Chlorophyceae	Cryptomonas	sp.	++	++	++
Bacillariophyceae	Detonula	confervaceae			++
Dinophyceae	Dinophysis	acuminata	+		+
Silicoflagellate	Distephanus	speculum		+	
Bacillariophyceae	Ditylum	brightwellii		+	+
Chlorophyceae	Dunaliella	sp.		+	
Bacillariophyceae	Euchampia	zoodiacus		+	++
Euglenophyceae	Euglena	sp.			+
Dinophyceae	Gonyaulax	rotundata			+
Dinophyceae	Gyrodinium	aureolum			+
Bacillariophyceae	Gyrosigma	sp.	++		
Bacillariophyceae	Leptocylindrus	danicus	++	++	+
Bacillariophyceae	Leptocylindrus	minimum	++		++
Bacillariophyceae	Melosira	nummuloides	+		+
Bacillariophyceae	Melosira	sulcata	++	+	+
Bacillariophyceae	Navicula	sp.	++	+	+
Bacillariophyceae	Nitzschia	closterium		+	+
Bacillariophyceae	Nitzschia	longissima	+	+	+
Bacillariophyceae	Nitzschia	seriata	+	+	+
Bacillariophyceae	Nitzschia	sp.	++		
Dinophyceae	Oxytoxum	sp.	++	+	
Prymnesiophyceae	Pavlova	sp.	++		
Dinophyceae	Peridinium	sp.		+	
Prymnesiophyceae	Phaeocystus	pouchettii	++		
Dinophyceae	Prorocentrum	micans		+	+
Dinophyceae	Prorocentrum	minimum		+	+

Continued on p. 310

Table 10. contd.

Class	Genus	Species	Hammonasset*	Milford	Stamford
Dinophyceae	Prorocentrum	scutellum	++	++	++
Dinophyceae	Prorocentrum	triestinum	++		++
Dinophyceae	Protoperidinium	sp.	+	+	
Prymnesiophyceae	Pyramimonas	sp.	+	+	
Bacillariophyceae	Rhizosolenia	delicatula		++	++
Bacillariophyceae	Rhizosolenia	faeroense		++	++
Bacillariophyceae	Rhizosolenia	fragilissima	++		+
Bacillariophyceae	Rhizosolenia	hebetata			+
Bacillariophyceae	Rhizosolenia	setigera		+	
Bacillariophyceae	Skeletonema	costatum	++	++	++
Bacillariophyceae	Thalassionema	nitzschioides		+	++
Bacillariophyceae	Thalassiosira	nordenskoldii	++	++	++
Bacillariophyceae	Thalassiosira	pseudonana	++	++	
Bacillariophyceae	Thalassiosira	rotula	++	++	+
Bacillariophyceae	Thalassiothrix	frauenfeldii	++	++	++
Total number of species by station, 1994			32	35	33
1995 Phytoplankton Species List					
Dinohyceae	Amphidinium	sp.			+
Bacillariophyceae	Amphiprora	sp.	+		
Bacillariophyceae	Asterionella	japonica		+	
Dinophyceae	Ceratium	sp.	+		
Bacillariophyceae	Chaetoceros	simplex			+
Bacillariophyceae	Coscinodiscus	sp.	+	+	++
Chlorophyceae	Cryptomonas	sp.	+	+	++
Bacillariophyceae	Detonula	confervaceae			++
Dinophyceae	Gonyaulax	rotundata		++	
Dinophyceae	Gymnodinium	sanuineum	+		
Bacillariophyceae	Gyrosigma	sp.	+		+
Dinophyceae	Katodinium	rotundatum		+	
Bacillariophyceae	Leptocylindrus	danicus	++	++	++
Bacillariophyceae	Leptocylindricus	minimum	++		
Bacillariophyceae	Melosira	sulcata	++	++	++
Bacillariophyceae	Navicula	sp.		+	
Bacillariophyceae	Nitzschia	sp.		+	+
Dinophyceae	Prorocentrum	scutellum			+
Dinophyceae	Protoperidinium	sp.	+	++	++
Prymnesiophyceae	Pyramimonas	sp.		++	
Bacillariophyceae	Rhizosolenia	faeroense	++		
Bacillariophyceae	Rhizosolenia	fragilissima	++	++	++
Bacillariophyceae	Skeletonema	costatum	++	++	++
Prymnesiophyceae	Tetraselmis	sp.			++
Bacillariophyceae	Thalassionema	nitzschioides	++	++	++
Bacillariophyceae	Thalassiosira	decipiens			++
Bacillariophyceae	Thalassiosira	nordenskoldii	+	++	++
Bacillariophyceae	Thalassiosira	pseudonana		++	++
Bacillariophyceae	Thalassiosira	rotula	++	++	++
Bacillariophyceae	Thalassiothrix	frauenfeldii	++		+
Total number of species by station, 1995			17	18	22

*Sampled only 6 months.

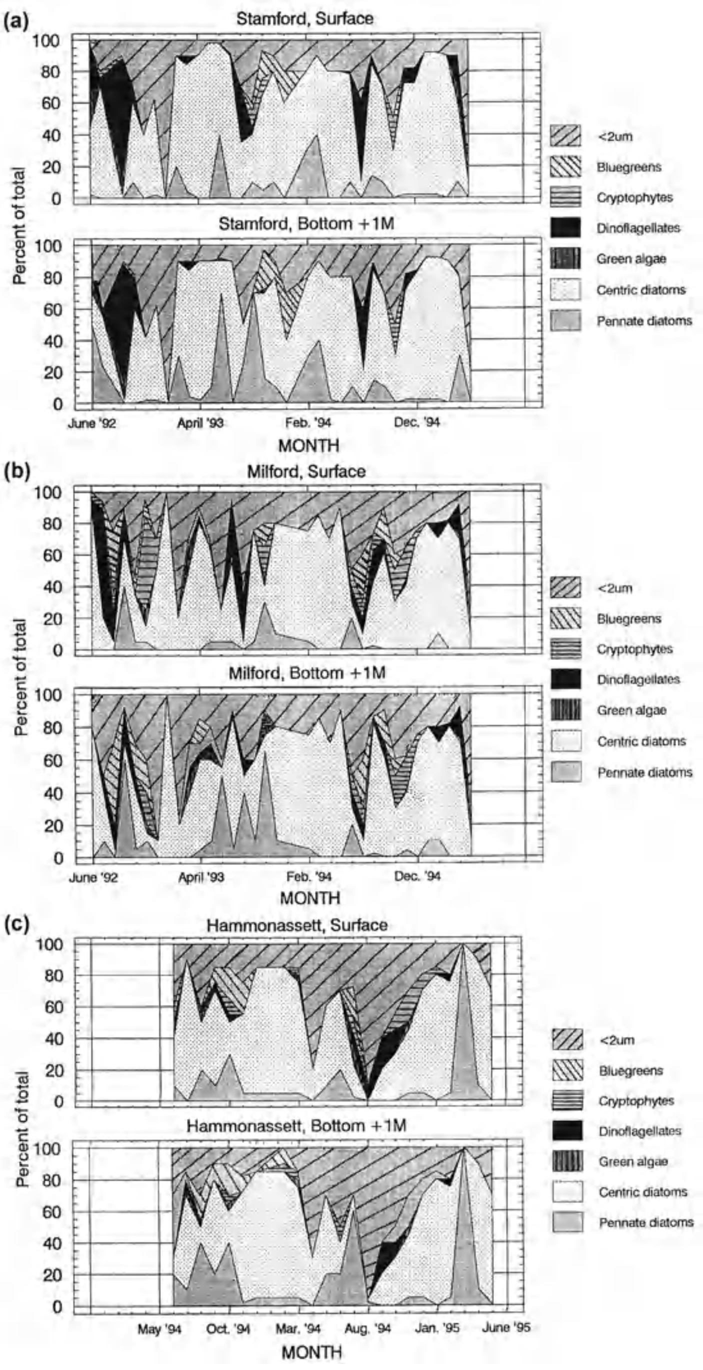

Figure 33. The percentage of phytoplankton assemblages in major taxonomic groups for the surface and bottom waters of all 3 study sites over the entire study period.

the western stations during spring, fall, and winter ($p \leq 0.05$). Milford ranked lowest during summer based on a nonparametric Kruskal–Wallis test (p=0.023), whereas Hammonasset ranked lowest the rest of the year (p=0.021).

In summary, the dominant pattern of biomass was an increase between Hammonasset and the western stations. The increase appeared especially strong in the spring, and it also prevailed in fall and winter. Gradients became variable and uncertain in the summer.

Table 11. Complete planktonic ciliate species composition list for the entire study period, for the 3 Long Island Sound study sites combined, based on protargol stained specimen analyses

Ciliate species list for all stations combined

Codonellopsis sp.
Codinella sp.
Cyrtostrombidium longisomum
C. wailesi
Didinium sp.
Euplotes sp.
Eutintinnus pectinis
E. lususundae
Favella arcuata
F. ehrenbergii
F. sp.
Haltera sp.
Helicostomella subulata
H. ssp.
Heterotricous condilostima
Laboea strobila
Leegardiella sol
Lohmanniella oviformis
Mesodinium sp.
Metacylis angulata
M. annulifera
Metastrombidium p.
Stenosemella oliva
S. steini
S. ventricosa
Strobilidium elegans
S. epacrum
S. marinum
S. multinucleatum
S. sphaericum
S. spiralis
S. undinum
S. veniliae
Strombidinopsis cheshira
S. multiauris
Strobidium acuminatum
S. acutum
S. basimorphum
S. bilobum
S. capitatum
S. compressum
S. conicum
S. daparedei
S. inclinatum
S. lynni
S. pelagicum
S. siculum
S. sulcatum
S. taylori
S. Titinnodes
S. tressum
S. ventropinnum
Tetraphyminid scutico
Tintinnopsis acuminata
T. baltica
T. beroidea
T. dadayi
T. fluviatile
T. kofoidi
T. levigata
T. minuta
T. parva
T. platensis
T. rapa
T. tubulosa
T. tubulosoides
T. urnula
T. vasculum
T. ventricosoides
Tontonia gracillima
T. poopsia

Table 12. Numbers and percentages of ciliate species encountered over the entire study period, for the entire study area, and by station, both as averages for the entire water column and by depth

Site	# of species	% of total	% in surface waters	% in bottom waters
Σ of all sites	71	100	–	–
Stamford site	67	94	72	69
Milford site	57	80	58	63
Hammonasset site	44	62	54	45

Species composition of the copepods

Results are presented for eight of the copepod species that were identified and enumerated. A ninth species will be mentioned in passing. All were observed at one time or another at each station (Table 15). Two major subgroups were noted; one comprising three species dominated the winter and spring seasons, and the other, with four species, dominated the summer and fall seasons. They typically overlapped briefly twice a year. From December through June (winter–spring season) *Acartia hudsonica* and *Temora longicornis* (Fig. 37) were predominant, and *Pseudocalanus* sp. was present in lower numbers (Fig. 38). *Centropages* sp. occurred during the spring and the summer (Fig. 38), but its numbers were low. From July through December (summer–fall season), *Acartia tonsa* usually dominated, and *Paracalanus crassirostris* could be abundant, especially during the fall (Fig. 39). There were two other summer–fall species, *Oithona* sp., and

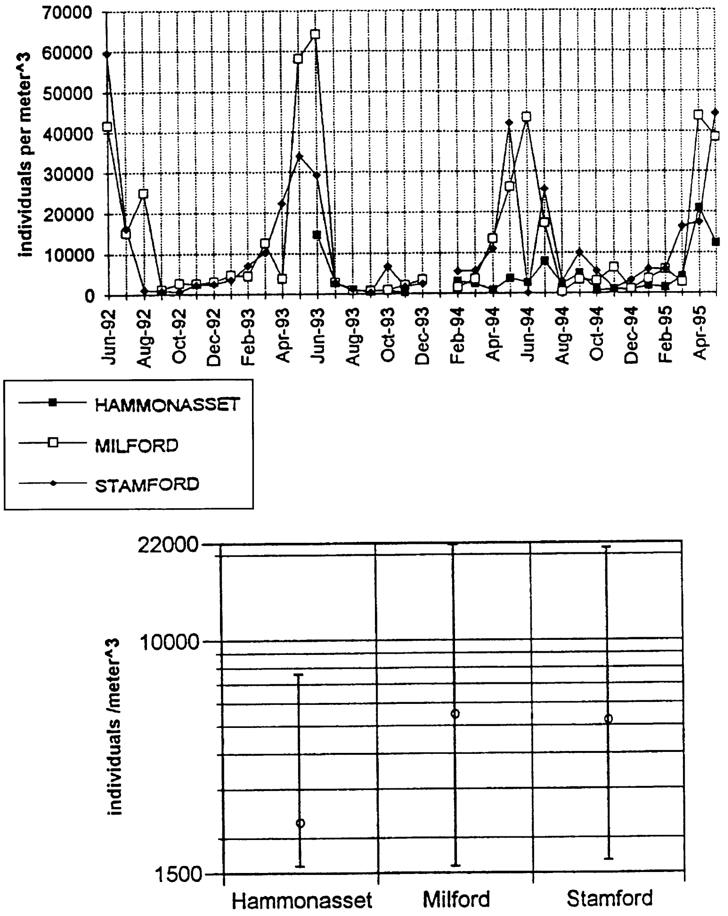

Figure 34. Number of copepods per m^3 (top), and the geometric means and standard deviations of copepod abundance (bottom) for the entire study period, averaged for the water column.

Labidocera sp. (Fig. 40), found in lesser numbers. A ninth species, *Eurytemora hardmanni*, was observed in insignificant numbers (2–4 copepods m^{-3}) in the spring. The seasonal species succession was well synchronized at all stations, and it was much the same from year-to-year for all of the dominant copepods (Fig. 41).

Patterns of the species among stations

Species composition was similar at each station, except that *Temora longicornis* was slightly more dominant at the two western stations during the winter and spring, and *Acartia tonsa* was slightly more dominant at the center station (Milford) in the summer and fall (Table 15). When all copepods were tallied over the period of study, 85% belonged to winter–spring varieties at Milford and Stamford, whereas the counts were somewhat more evenly divided between winter–spring and summer–fall varieties at Hammonasset (Table 15). One genus, *Centropages* sp., actually decreased from east to west, but this was one of the less abundant copepods. *Centropages typicus* and *Centropages hamatus* were considered by Deevey (1956) as primarily neritic (or less tolerant of reduced salinity), possibly explaining their distribution.

In summary, nine species were identified and counted, and all were present at each station. There were two major subgroups, winter–spring (boreal), and summer–fall (warm water). In winter and spring, *Temora longicornis* and *Acartia hudsonica* were numerically dominant. In summer and fall, *Acartia tonsa* and *Paracalanus crassirostris* were numerically dominant. There was no striking difference among stations in the species compostion of the copepod

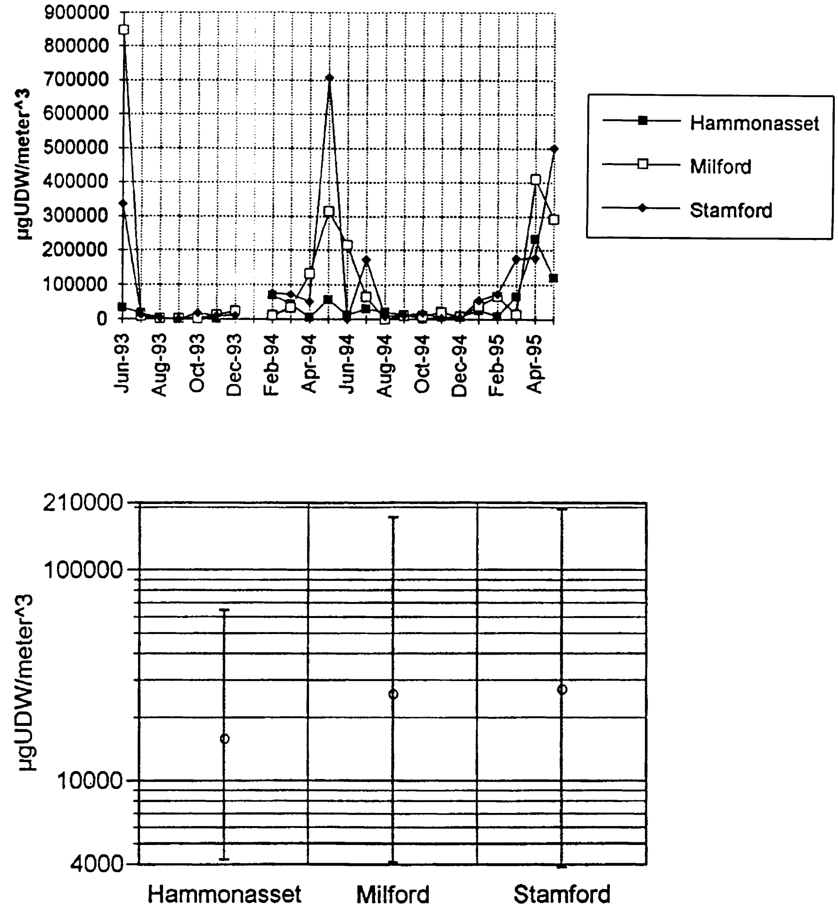

Figure 35. Combined biomass of the copepods *Temora longicornis,/ Acartia hudsonica* and *Acartia tonsa* (top), and the geometric means and standard deviations of biomass (bottom) for the entire period of study (values are water column integrated averages), presented as unpreserved dry weight (UDW).

Table 13. Descriptive statistics of copepod abundance for the period of the study

	Numbers of individuals per m^3		
	Hammonasset	Milford	Stamford
Mean	4162	13 288	11 677
Geometric mean	2303	5548	5282
Minimum	361	415	183
Maximum	21 053	64 152	59 542
Standard deviation around the mean	±5301	±17 793	±14 535
Standard deviation around the geometric mean	+5527 −717	+16 065 −3967	+15 721 −3620

Table 14. Descriptive statistics for the combined biomass of 3 major copepod species, *Temora longicornis, Acartia hudsonica*, and *Acartia tonsa*, over the period of study in units of unpreserved dry weight (UDW)

	μg UDW/m^3		
	Hammonasset	Milford	Stamford
Mean	36 217	110 715	109 229
Geometric mean	16 103	26 213	27 535
Minimum	1 765	1 036	405
Maximum	223 775	847 554	707 655
Standard deviation around the mean	±53 180	±198 365	±179 214
Standard deviation around the geometric mean	+48 546 −11 903	+152 597 −22 123	+168 490 −23 671

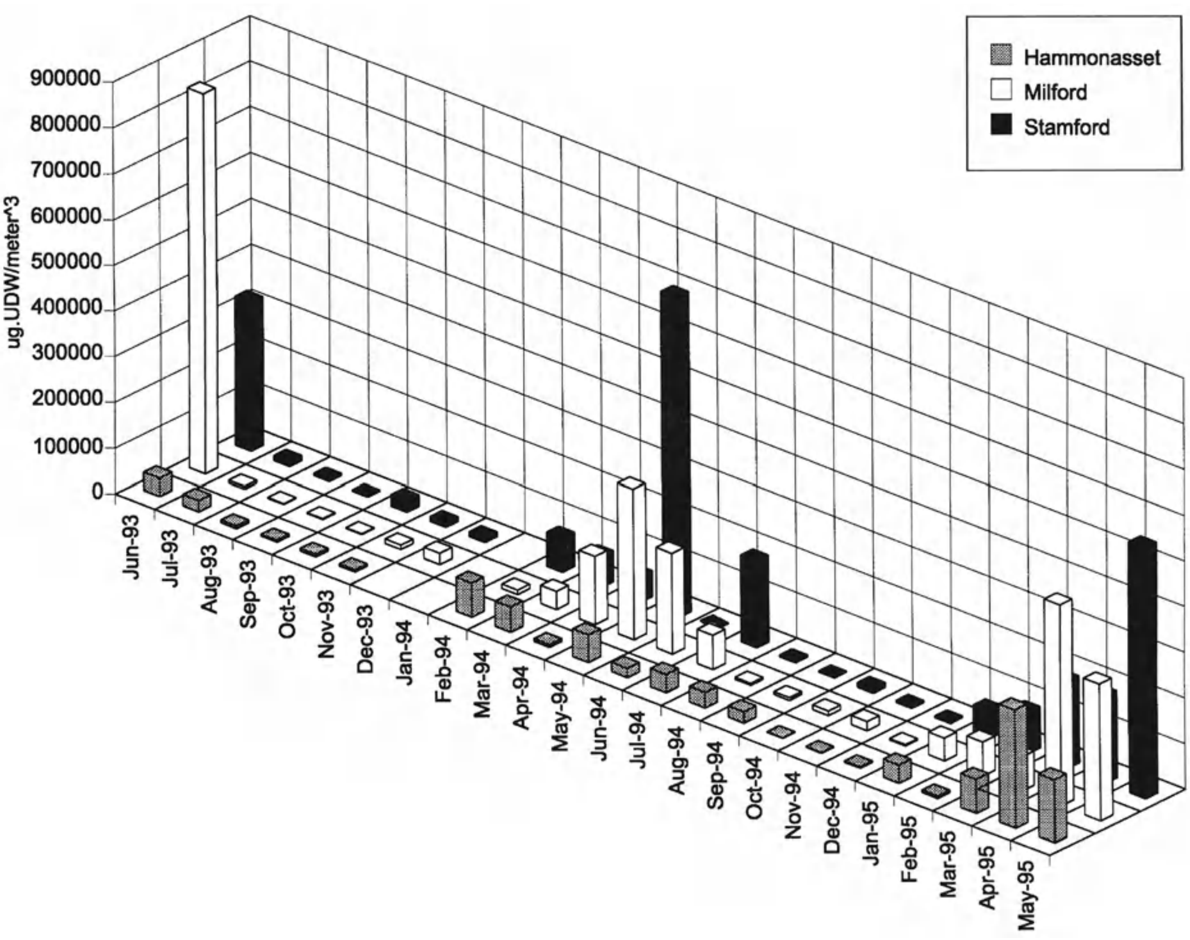

Figure 36. Copepod biomass, as μg unpreserved dry weight (UDW)/m^3) by station. In the entire period of study (values are water column integrated averages).

community. However, over the period of study most of the copepods in the west occurred in the winter–spring assemblage due to massive springtime populations, while at Hammonasset they were more evenly divided between winter–spring and summer–fall assemblages.

Correlations between copepod data & other variables

Seasonal patterns of copepod abundance and (or) biomass correlated positively with Chl and (or) Chl >20 at all three stations. Moreover, stations with more chlorophyll and Chl >20 had more copepods, thus copepods and chlorophyll appeared to associate spatially with one another along the eutrophication gradient. There were no correlations between copepods and Chl 10–20 or Chl <10. Copepod numbers and (or) biomass were correlated negatively with DIN and NO_X concentrations at two of three stations, which was due to very low NO_X concentrations during the peaks in copepods. The overall increase in copepods took place between Hammonasset and Milford, whereas a small increase in DIN occurred between Milford and Stamford, thus there was no strong spatial association between DIN concentration and copepods along the eutrophication gradient. Chl and Chl >20 concentrations were negatively correlated with DIN and NO_X concentrations, and positively correlated with PN concentrations, suggesting the uptake of DIN by algae and conversion to PN.

Dissolved inorganic nitrogen concentrations were negatively correlated with PN and DON concentrations, implying seasonal trade-offs between inorganic and organic forms of nitrogen. Correlations of TDN concentrations with DIN concentrations as well as DON concentrations suggest that control of TDN levels can lie with either DIN or DON. Correlations of TN levels with TDN concentrations as well as PN con-

Table 15. Means of copepod abundance by species in units of ind/m^3; relative proportions of each species, as pecentage, for winter–spring, or summer–fall copepod populations, and relative proportion of all the copepods, counted over the period of study, occurring in the winter–spring and summer–fall subgroups

	Hammonasset		Milford		Stamford	
	X	%	X	%	X	%
Winter–Spring Copepod Group						
Temora longicornis	868	29	4431	40	3623	37
Acartia hudsonica	2070	69	6422	59	5972	62
Pseudocalanus sp.	61	2	117	1	91	1
Summer–Fall Copepod Group						
Acartia tonsa	895	74	1680	85	1438	74
Paracalanus sp.	287	24	245	12	475	24
Oithona sp.	17	1.5	35	2.5	33	1.7
Labidocera sp.	8	0.5	8	0.5	5	0.3
A Genus Present Mostly in Spring–Summer						
Centropages sp.	89		56		14	
Relative Proportions of the Winter–Spring and Summer–Fall Groups within the Overall Population						
Winter–Spring		70		85		84
Summer–Fall		30		15		16

centrations suggest that control of TN levels is shared between dissolved and particulate forms of nitrogen.

Copepods are an important part of the food web in LIS, and their seasonal and spatial distributions can be influenced by the quantity and quality of phytoplankton, as well as by water temperature and predation. The distribution of phytoplankton may in turn be influenced by the quantity and chemical composition of dissolved nitrogen, among other things. Therefore, a eutrophication gradient in LIS appears to influence the spatial distribution of copepod populations.

Zooplankton other than copepods

The zooplankton focused on in this study were the protozooplankton and the microcrustaceans (i.e. the copepods). We did, however record all non-copepod macrozooplankton encountered in our sub-samples. Since our sub-sampling was designed to quantify the copepods, we often encountered too few 'other zooplankton' to come up with reliable quantitative estimates. We did, nonetheless, note their presence or absence as ecological functional groups. Some seasonal patterns appear in certain of the larval groups and in the gelatinous depris, which is most common in the summer and fall sampling periods. Of particular noteworthness is the fact that gelatinous zooplankton wet volumes, primarily derived from ctenophores bodies, appears to be much more prevelant at our western Long Island Sound Stamford station.

Larval fish species composition & densities

In all, over the 3 years of study (1992–1995), 18 species of larval fish were encountered and ranked according to abundance, at our 3 Long Island Sound inshore stations (Table 16). The number of species encountered on any sampling date varied from 0 to 6 (Figs 42 and 43) with densities varying from 0 to a high of 120 per cubic meter (Figs 44 and 45). Similar trends in diversity as well as abundances were observed at all 3 stations. When differences were observed they typically were minimal, with Stamford, CT waters having higher concentrations. The 18 species encountered in this study compares to 22 species observed by S. Richards (Riley, 1955; Riley et al., 1956) in her 1952–1955 data set. Comparison of the 1950s and 1990s data indicates that 15 larval fish species were common to both data sets. Three species were found in the present study but not in the 1950s samples (Table 17), and 7 species were observed in the 1950s samples but not in our 1990s data set (Table 18). It is of particular interest that most of the species missing in the current study are species that have either experienced intensive fishing pressure (e.g. 4-Spotted and Yellowtail Flounder, Sea Robins), predator-induced mortality from estuary-feeding birds such as cormorants, or hypoxia stress (e.g. Atlantic Silverside).

Discussion

The various agents of eutrophication affecting Long Island Sound have only slightly elevated levels of dissolved nitrogen compounds, and more significantly enhanced dissolved phosphate levels in Long Island Sound (primarily in the western portions of the estuary. Nitrate and nitrite concentrations throughout the Sound are similar in concentration to those reported in the 1950s by Riley (1955) and Riley et al. (1956). The relative proportioning among chemical species of nitrogen, however, differs from west to east, both contemporaneously, as well as temporally, with NH_4 and dissolved organic nitrogen at times more prevalent in the west (particularly in bottom waters). The excess loadings of nitrogen, and other nutrients into the western Sound appear to be taken up by the planktonic

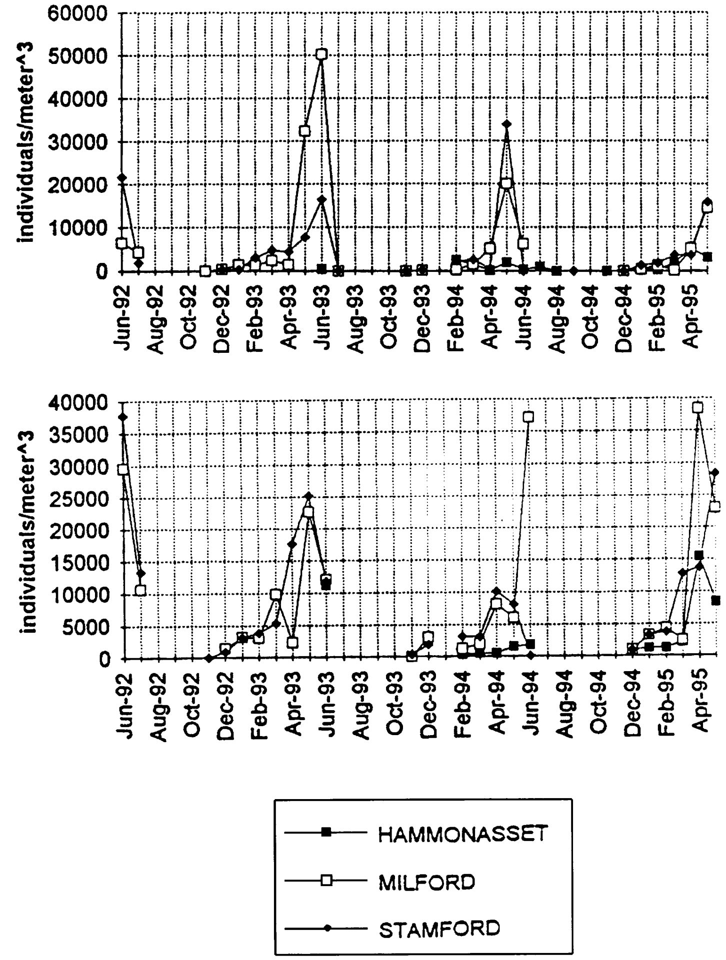

Figure 37. Abundance of individual copepods of the species *Temora longicornis* (top) and *Acartia hudsonica* (bottom), for the entire study period (values are water column integrated averages).

food web and converted to biomass as evidenced by observed bacterial, chlorophyll, phytoplanktonic and zooplanktonic west to east concentration gradients. Size-fractionated chlorophyll data indicate little east to west differences in the 10–20 μm size fraction, while pointing to large differences in the <10 and >20 μm fractions, which are both higher in the west. Occurring along with the enhanced phytoplankton biomass is slightly enhanced bacterial densities and growth rates. The densities show interesting seasonal cycles and appear to be related not to total chlorophyll levels but to densities of the photosynthetic nanoplankton. Heterotrophic nanoplankton densities are also higher in the west and appear also to at times influence bacterial densities.

Species composition of phytoplankton routinely differ among west to east stations. These species compositional shifts appear to be related to N/P and N/Si ratios, as well as to ratios among nitrogen chemical species. Dissolved inorganic N/P ratios are routinely low among all stations , with the west exhibiting lower levels than the east. However, total dissolved N/P ratios (which include dissolved organic nitrogen) are similar among stations and typically are well above the Redfield ratio of 16:1.

Associated with enhancement of bacterial, HNAN and <10 μm chlorophyll is significant enhancement of planktonic ciliate species diversity in the western Long Island Sound waters.

Microcrustacean (i.e. copepod) biomass is extremely enhanced in the west *versus* the east, indicating that while stimulating the microbial loop, eutrophication is also enhancing secondary production preferred by larval fish, comb jellies and jellyfish.

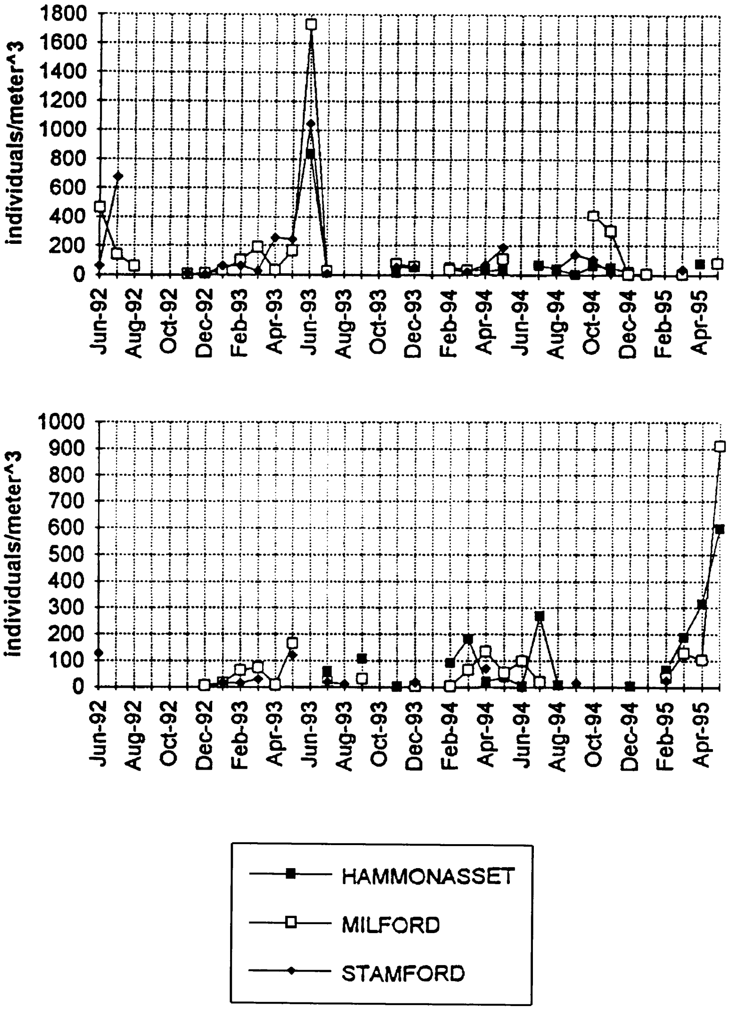

Figure 38. Abundance of individual copepods of the species *Pseudocalanus* sp. (top) and *Centropages* sp. (bottom); for the entire study period (values are water column integrated averages).

Copepod biomass as well as fecal pellet production likely is a significant contributor to hypoxia formation in western Long Island Sound. Copepods increased in number and biomass between the eastern station at Hammonasset, and the two western stations at Milford and Stamford, particularly in the spring. In addition, the spatial pattern of copepod stocks matched best with the spatial pattern of large-size phytoplankton (Chl >20), as opposed to any of the other chlorophyll or nitrogen parameters that were measured. Moreover, the seasonal cycles of copepods, chlorophyll, and nitrogen were in good general agreement with the overall scheme for LIS in the literature, with certain exceptions: (1) High winter NO_X levels lasted longer, until March or even April, although peaks were similar in height. (2) Flowerings during the winter–spring period peaked in March or April, rather than February or March. (3) Chlorophyll levels were higher on average, and particularly so in summer. (4) Spring peaks in copepods were lower than expected at Hammonasset. (5) The summer–fall copepods did not achieve population levels as high as expected. These differences could be due to offshore-inshore gradients, and (or) spatio-temporal changes brought about by eutrophication. Copepod abundance and biomass increased between the eastern station and the central station, but not between the central station and the western station. Events in the temperature cycle, the copepod abundance cycle, and the species succession occurred simultaneously at all three stations, which argues that temperature affected the copepods seasonally, but not spatially. The greatest spatial increases in copepods

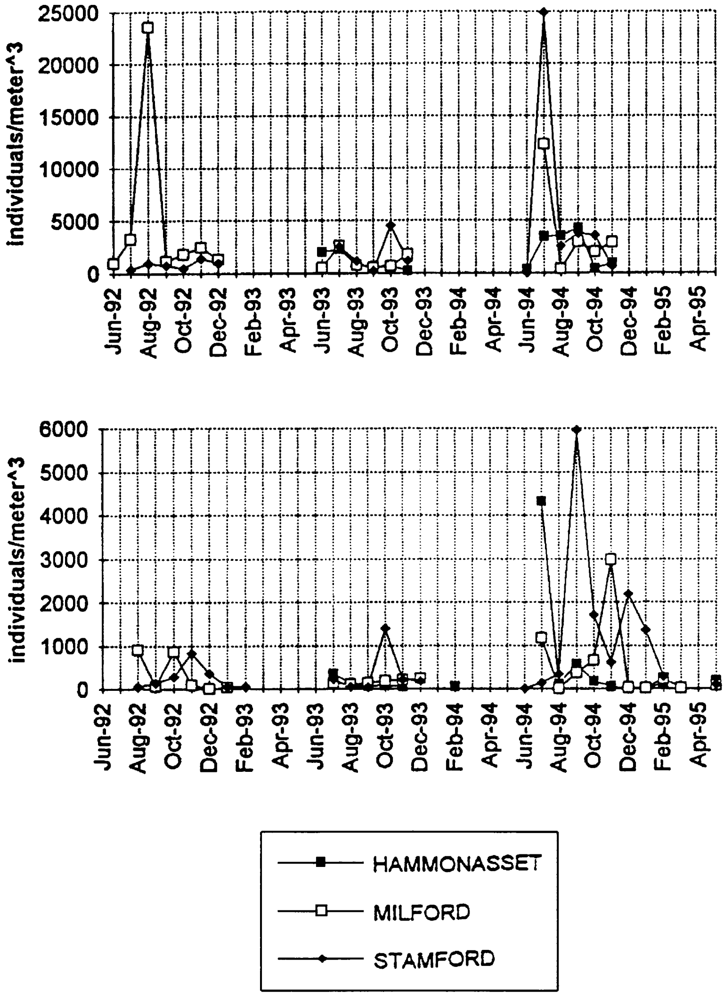

Figure 39. Abundance of individual copepods of the species *Acartia tonsa* (top) and *Paracalanus crassirostris* (bottom) for the entire study period (value are water column integrated averages).

took place in the spring, and a difference in the food supply among stations could be responsible.

Yearly differences in the characteristics of the peak in winter–spring copepods seem to correspond to characteristics of the winter–spring bloom (Harris & Riley, 1956; Harris, 1959; Dam, 1989). This leads to suspicion that spatial differences in the characteristics of the blooms might correspond to spatial differences in the copepods, and there is some evidence of this. Peak populations of winter–spring copepods followed peak concentrations of Chl >20, and peaks in copepods as well as in Chl >20 lasted longer at Milford and Stamford than they did at Hammonasset (Fig. 46). Moreover, the east–west trends in copepod standing stocks and the east–west trends in Chl >20 were similar in size and direction (Fig. 47). In addition, copepod standing stocks and Chl >20 were significantly correlated to each other at all stations.

Strong patterns of east–west increase in the abundance of the boreal copepods gave way in summer to much weaker east–west patterns in the abundance of the warm-water copepods. Conover (1956) suggests that the seasonal switches between boreal and warm-water copepods involve temperature dependent competitive interactions of special importance to fecundity and juvenile growth, more than lethal effects. Boreal species receded rapidly when temperatures increased to 18–20 °C, and the tropical species gave way more slowly when temperatures decreased again to 15–10 °C. The peak populations of winter–spring copepods in May and June collapsed, as expected, when water temperatures began to exceed 18 °C, but the summer–

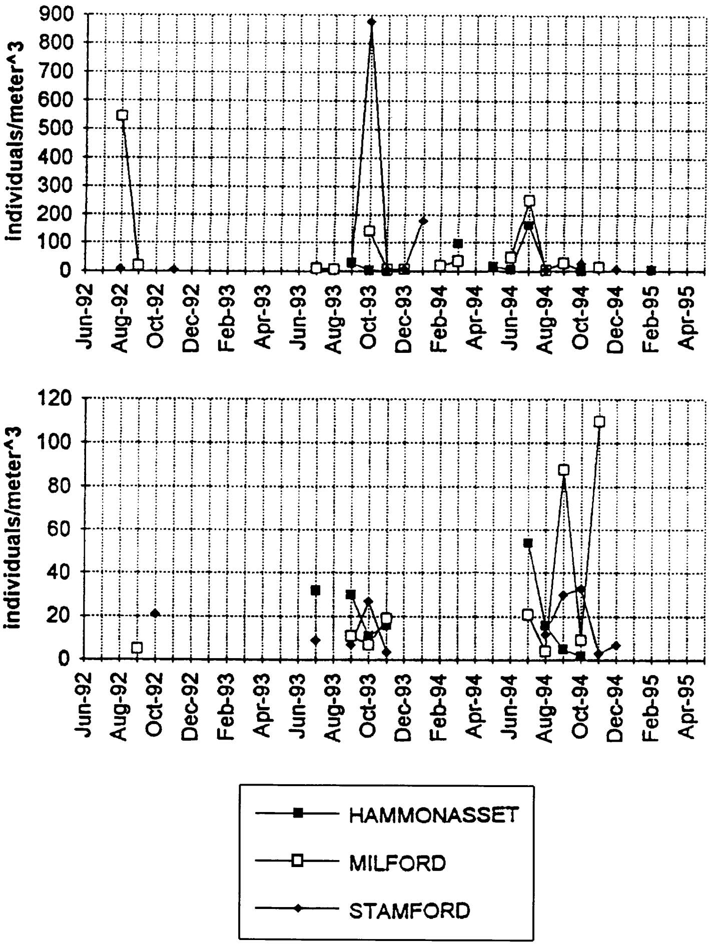

Figure 40. Abundance of individual copepods of the species *Oithona* sp. (top) and *Labidocera* sp. (bottom) for the entire study period (values are water column integrated averages).

fall varieties replacing them never achieved similarly high population levels. Copepod stocks were uniformly low in summer and fall, with only two notable exceptions. One exception was in July 1994, when copepods peaked at all three stations, and the other was in August 1992 only at Milford. Copepods did not appear to be food limited (based on our unpublished data and a food limitation model not presented here) in summer on an assumed diet of Chl >20, and as mentioned above, there is little question that the likelihood of food limitation was reduced even further by omnivory.

Circumstantial evidence implicates ctenophores in controlling the abundance of the summer–fall copepods. In summer and fall of 1992, 58% of zooplankton samples held ctenophore debris (ctenophores preserve very poorly in formalin). In summer and fall of 1993, 45% of zooplankton samples held ctenophore debris. In summer and fall of 1994, however, ctenophore debris occurred in only 27% of zooplankton samples, and 1994 saw the occurrence of a peak in standing stocks of summer–fall copepods at all three stations. Predation rates of 20–100% d^{-1} by seasonal ctenophore swarms on copepods have been noted to occur in various northeast estuaries (Bishop, 1967; Kremer, 1979; Johnson, 1987). Johnson (1987) also noted an inverse relationship between numbers of ctenophores and numbers of copepods.

The east–west trends of increase in nitrogen concentrations were much weaker than the east–west trends of increase in both chlorophyll concentrations and copepod abundance. The DIN fraction was usually slightly more concentrated west of Hammonasset in the winter, which may have had something to do with the east–west patterns of chlorophyll and copepods in the spring. Nitrogen loading and primary produc-

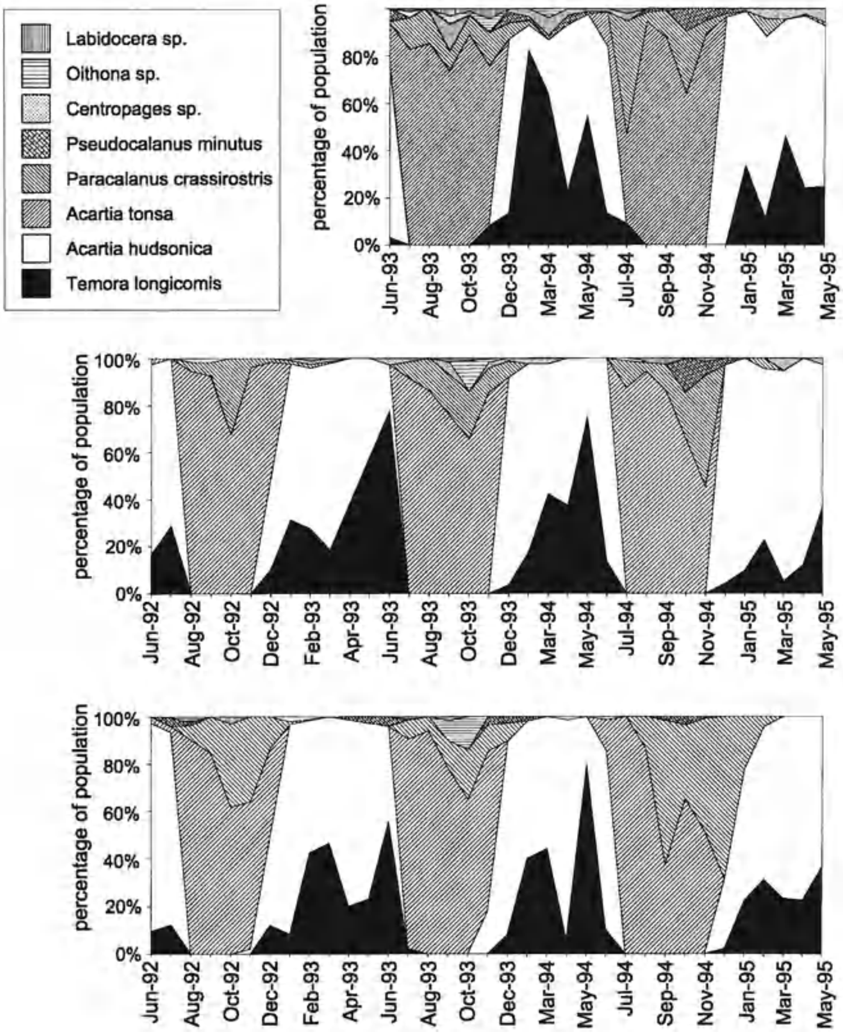

Figure 41. Species composition of the water column copepod populations at the Hammonasset (top), Milford (center) and Stamford (bottom) stations for the entire period of study.

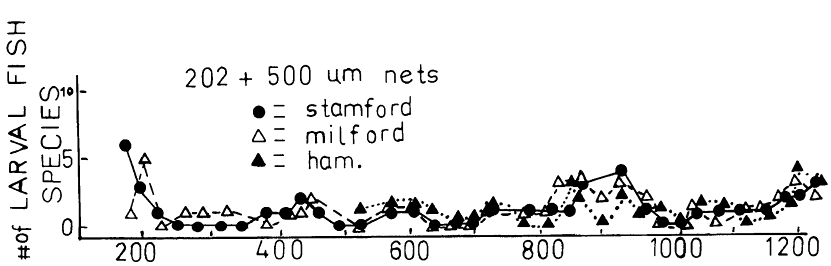

Figure 42. Number of larval fish species encountered at each of the 3 stations for the 202 μm and 500 μm combined water column net samples, over the entire study period (expressed as Julian days, commencing on January 1, 1992).

Table 16. Larval fish species encountered in our inshore Long Island Sound waters, and the abundance ranking for each (1992 – 1995)

Common name	Larval Fish Species Encountered 1992–1995 Inshore Long Island Sound Waters Genus & species	Abundance ranking
Wry Mouth	*Cryptocanthodes maculatus*	12
Butterfish	*Peprilus triacanthus*	10
Pipefish	*Syngnathus fuscus*	10
Windowpan Flounder	*Scophthalmus aquosus*	9
Four-bearded Rockling	*Enchlopus cimbrius*	8
18-spine Sculpin	*Myoxocephalus octodecemspinosus*	7
Tautog (Blackfish)	*Tautoga onitis*	6
Menhaden	*Brevoortia tyrannus*	5
Cunner	*Tautogolabrus adspersus*	4
Winter Flounder	*Pseudopleuronectes americanus*	3
Sand Lance	*Ammodytes americanus*	2
Anchovy	*Anchoa mitchilli*	1
Herring	*Clupea harengus*	Occasional
Cod	*Gadus morhua*	Occasional
Little Sculpin	*Myoxocephalus aeneus*	Occasional
Scup (Porgy)	*Stenotomus chrysops*	Occasional
Hogchocker	*Trinectes maculatus*	Occasional
Sea Snail	*Liparis liparis*	Occasional

Table 17. Larval fish species encountered in the present study (1992–1995), but not found in S. Richards (Riley et al., 1956, 1959) 1952–1955 work

Common name	Larval Fish Species Encountred in the Present Study (1992–1995) but not found in S. Richards (Riley et al. 1955) 1952–1955 work Genus & species
Wry Mouth	*Cryptocanthodes maculatus*
Hogchocker	*Trinectes maculatus*
Sea Snail	*Liparus liparus*

Table 18. Larval fish species encountered in S. Richards (Riley et al., 1956, 1959) work for 1952–1955, but not found in the present study (1992–1995)

Common name	Larval Fish Species Encountered by S. Richards (Riley et. al., 1955) for 1952–1955, but not Found in the Present 1992–1995 Study Genus & species
Atlantic Silverside	*Menidia menidia (notata)*
Northern Kingfish	*Menticirrhus saxatilis*
Northern Sea Robin	*Prionotus carolinus*
Striped Sea Robin	*Prionotus evolans*
4-Spotted Flounder	*Paralichthys oblongus*
Yellowtail Flounder	*Limanda ferruginea*
Northern Puffer	*Sphaeroides maculatus*

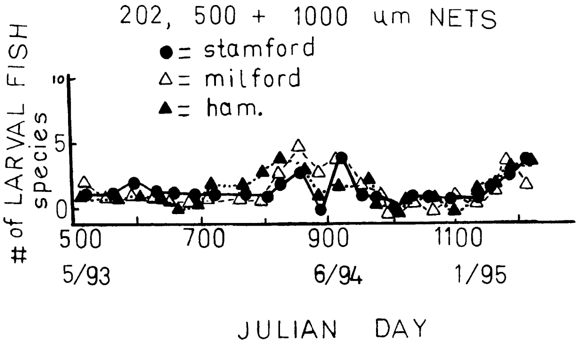

Figure 43. Number of larval fish species encountered at each of the 3 stations for the 202 μm, 500 μm and 1000 μm combined water column net samples, over the entire study period (expressed as Julian days).

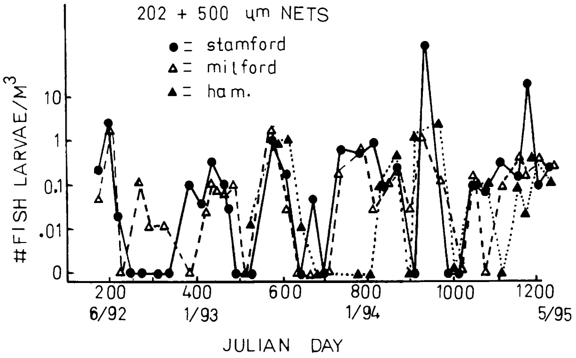

Figure 44. Number of larval fish/m^3 at each of the 3 stations for the 202 μm and 500 μm combined water column net samples, over the entire study period (expressed as Julian days).

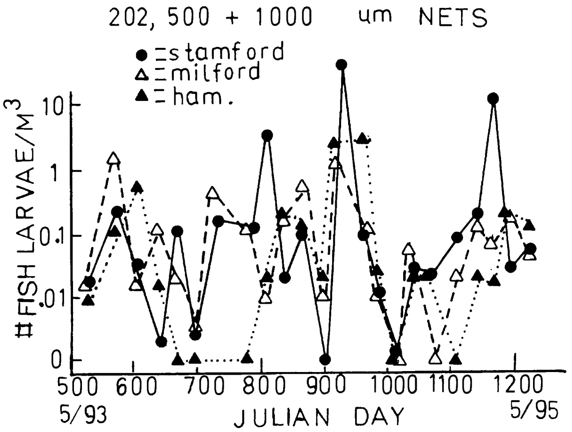

Figure 45. Number of larval fish/m3 at each of the 3 stations for the 202 μm, 500 μm, and 1000 μm combined water column net samples, over the entire study period (expressed as Julian days).

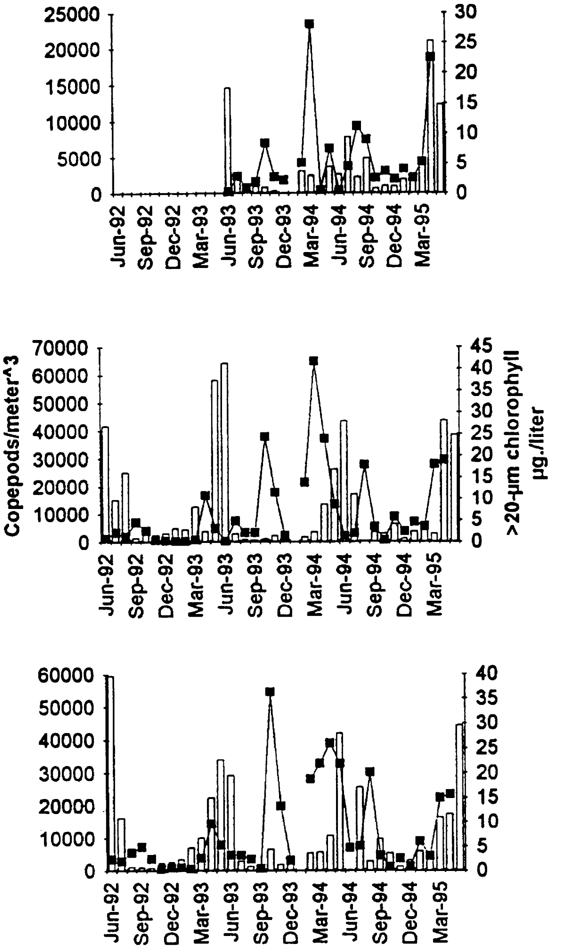

Figure 46. Copepod abundance (as histograms) and depth-averaged greater than 20 μm chlorophyll (line graphs), at the Hammonasset (top), Milford (center), and Stamford (bottom) stations.

tion were well correlated in three of four estuaries studied by de Jonge et al. (1995), including LIS; leading one to conjecture that primary production rates may act to buffer nitrogen concentrations in such a way that regional differences in nitrogen loading do not cause proportional regional differences in nitrogen concentration. It could also be suggested that grazers buffer chlorophyll concentrations, only not as well as primary production buffers nitrogen; perhaps explaining why small east–west increases in nitrogen were associated with larger increases in chlorophyll, and even larger increases in copepods. The occurence of top-down-control over standing stocks in food webs has been noted (see reviews by Carpenter et al., 1985; McQueen et al., 1989; Hunter & Price, 1992; Power, 1992; Strong, 1992). Thus, the large east–west increase in the winter–spring copepod population is

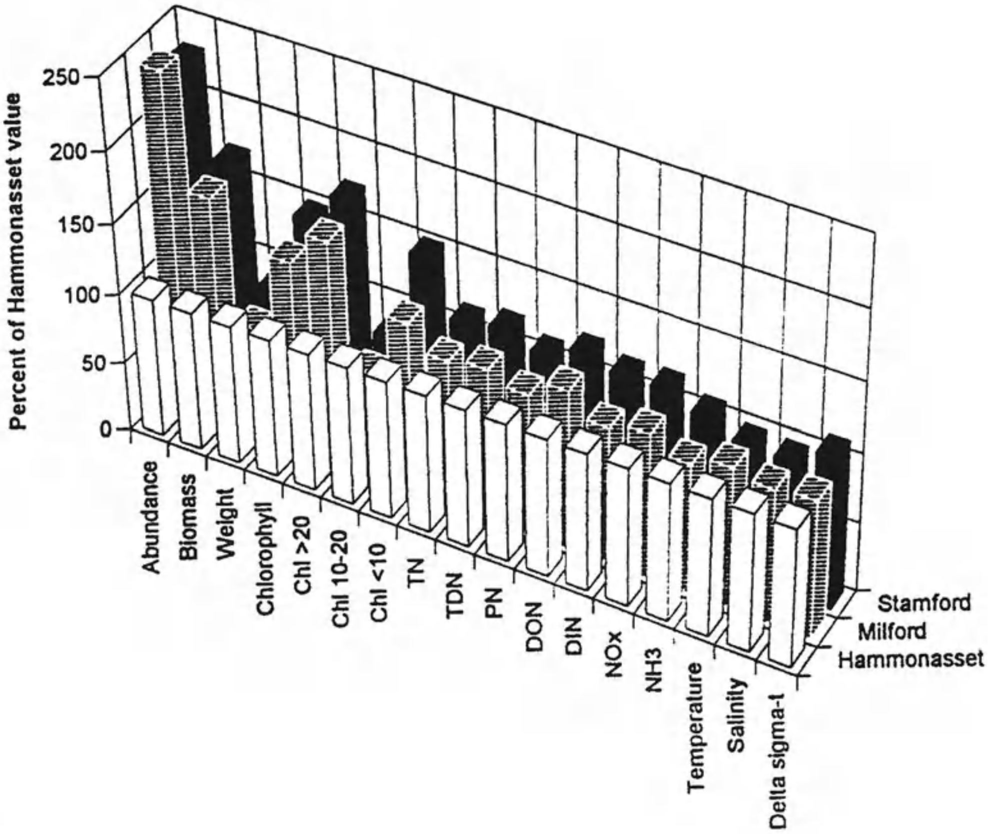

Figure 47. Trends among stations in copepod standing stock and individual weight, and trends among stations in chlorophyll concentrations, nitrogen concentrations and 3 water column physical properties.

best explained as a product of the east–west increase in the availability of large-sized phytoplankton. The summer–fall copepods, on the other hand, may have been limited by predation more than food, which may have led to their lesser abundance, and the greater uniformity of their east–west distribution.

Eutrophication can be thought of as a syndrome (suite of symptoms) that changes with the seasons. For example, the foremost symptom of eutrophication before a bloom might be abnormally high levels of dissolved nitrogen; whereas the symptoms during the bloom would become abnormally high chlorophyll concentrations and primary production maintained over an abnormally long period of time. After the bloom, there could be an abnormally high and persistent amount of herbivorous zooplankton biomass and production, and/or an increased amount of organic matter awaiting decomposition. Other symptoms of eutrophication include increased oxygen depletion below the summer thermocline (Welsh & Eller, 1991), and increased turbidity (Lund, 1969).

The eutrophication gradient, as indicated by the chlorophyll and nitrogen data, occurred primarily between Hammonasset and Milford. There was little difference between Milford and Stamford. This situation might reflect the fact that Milford is located about 5 k east of the mouth of the Housatonic river, the second largest river entering LIS, and a source of nitrogen. Therefore, the east–west gradient along the CT shore of LIS may be interrupted by this source. Hammonasset is located about 15 kilometers west of the mouth of the Connecticut River, which is the largest source of riverine freshwater to LIS. The plume of the CT river tends to the east, towards the nearby connection of LIS with the Atlantic Ocean (Fig. 1); moreover, Hammonasset is in a region where the net flow tends to be towards the east near-shore (O'Donnell, personal communication about drifter and current meter studies, and Welsh, personal communication about bathymetry and circulation in LIS). All of which suggests that nitrogen from the CT river might not as directly affect events at the Hammonasset station. Riley &

Conover (1956) suggested a mechanism whereby nutrients would be conserved and accumulated in LIS provided (1) residual circulation of water fits the classical estuarine pattern of up estuary (west) at the bottom, and down estuary (east) at the surface, (2) algal growth produces a vertical gradient of nutrients, and (3) a proportion of the algae produced at the surface is removed to deeper waters. Circulation in LIS appears to fit the classical estuarine circulation pattern in the eastern basin, but elsewhere it becomes more complex (Valle-Levinson & Wilson, 1984).

Nitrogen loading to LIS has more than doubled since colonial times, and about 50% of the nitrogen entering LIS comes from sewage treatment plants, which are clustered towards the western end (EPA, 1994; Welsh et al., 1995). Moreover, the East River, which connects western LIS to the waters surrounding New York City, is the single largest source of anthropogenic nitrogen. Increase in the sewage load may have leveled off since colonial times, but the form of the dissolved nitrogen fraction has shifted towards inorganic forms since the general conversion to secondary sewage treatment. The conversion is likely to have caused an increase in primary production because DIN is taken up directly by phytoplankton, whereas DON tends to require remineralization before usage. Increases in decomposition and biological oxygen demand below the thermocline are associated with increases in primary production, which may have caused an expansion of dangerously low oxygen levels in recent decades (Welsh et al., 1995). In short, the east–west gradient persists, and an increased inorganic nitrogen load contributes to the growth of phytoplankton, which might explain part of the difference between Riley & Conover's (1956) chlorophyll data and the data in this study.

The spatial variations in chlorophyll associated with eutrophication appear even larger than the seasonal variations if the western extremity of the sound (not sampled in this study) is included. Riley (1959) noted a 700% average increase in chlorophyll between the eastern and western extremes of LIS, reflecting a sharp upturn near the East River (New York City). Riley (1959) shows an increase an order of magnitude lower between the approximate longitudes of Hammonasset and Stamford, nearer the 50% estimate of this study. East to west increases in DIN are more subtle; they appear small or nonexistent in summer, but become more obvious after August (Riley & Conover, 1956). East to west increases in zooplankton did not seem to be clearly significant to Riley (1959) based on the data he had available. Since most of Riley's stations were further offshore than the stations used for this study, there is a chance that the significant east–west increase in copepods along the CT shore may not be typical of deep water areas in LIS.

Clearly, differences in prevailing conditions between inshore and offshore waters may affect the interpretation of these results as opposed to those of Riley and his group with respect to eutrophication and copepods in LIS as a whole. Increased concentrations of phytoplankton and zooplankton are noted by Riley (1955) in the shallow waters along the CT shore, thus eutrophication in LIS has a cross-Sound or south to north aspect as well as an east to west aspect. Hardy (1970) shows a distinct south to north increase of DIN concentrations in the central basin of LIS, which could be the root cause of the inshore-offshore gradients in phytoplankton and zooplankton Riley noted. Nearshore gradients in tidal mixing (Peterson, 1986), and influxes of fresh water (Riley, 1956) provide nutrients and alter the balance between stability and turbulence (Margalef, 1978; Schnitzer, 1979; Demers et al., 1986; Legendre & Lefévre, 1989). The general paradigm that small dinoflagellates replace large diatoms during the summer due to warmth, stratification, and low DIN is equivocal near shore, or any time there is a slight increase in NH_3 (Riley, 1969; Peterson, 1986). Inshore–offshore contrasts probably account for some of the difference between chlorophyll concentrations observed in this study, and those Riley (1959) shows at stations further offshore.

Processes considered

Broad generalities linking copepods and eutrophication are difficult to find because each system is different. In Osaka Bay and Tokyo Bay during summer, the median copepod weight decreased along a gradient of increasing eutrophication. Copepod biomass also decreased with eutrophication in Osaka Bay, but not in Tokyo Bay. Moreover, the species of copepods were shifted towards smaller varieties over the past several decades as eutrophication increased, and microphytoplankton gave way to picophytoplankton and nanophytoplankton (Uye, 1994). Why did Uye note reduced copepod biomass with eutrophication and we did not? Uye suggests, with good reason, that the displacement of large phytoplankton by small phytoplankton affected the copepods. In this study, copepod abundance, copepod biomass, and Chl >20 increased spatially with eutrophication. Thus it seems a paradox that average copepod weight decreased with eutrophication in this study as well as Uye's. The copepods in LIS were 10–20% lighter at the stations with the most Chl >20, a finding for which we presently have no good explanation. Predators might be picking off more of the large copepods in the west than the east, and there is some evidence that larval fish may be more abundant at the western stations.

The next example is similar to what was observed in LIS, at least in the relation between copepods and phytoplankton. As water in the vicinity of the Southern Benguela upwelling aged and lost its charge of DIN, phytoplankton stocks decreased and dominance shifted from the size fraction >10 μm to the size fraction <10 μm. Moreover, copepods were fewer where phytoplankton <10 μm were dominant (Walker & Peterson, 1991; Painting et al., 1993). Copepod abundance and biomass gradients appear to occur where there are meaningful differences in productivity and cell size distribution. Extended flowerings appear more conducive to large populations of copepods than short blooms; moreover, the presence of a steady supply of food particles that are large enough seems to be a prerequisite. It would seem that some enrichment is required for large copepod populations, but that eutrophication beyond a certain point could be bad for them. Turbidity may become raised to such a level that the ability of flagellates to hold position in a narrowed euphotic zone allows them to displace the large phytoplankton that generally favor copepods. Perhaps the ratio of nitrogen – silicon becomes too high to allow diatoms to compete. Bottom water anoxia caused by eutrophication has also been implicated as a cause of change within the copepod community, with some species living associated with the benthos for parts of their life cycle (Purcell, 1994; Uye, 1994). The literature provides less detail on the seasonal behavior of DON than for DIN and PN, but the importance of DON as a bacterial substrate and source of recycled DIN is an accepted fact, and is supported by data from the larger study indicating large and consistent summer bacterial peaks. Our estimate that DON accounts for about 40% of TN in LIS on average is somewhat lower than the estimate (50–64%) of Sharp et al. (1982) for marine systems in general. Harris (1959) reported low DON values one spring, and there was similar evidence in this study, but neither one provided good evidence for a consistent yearly cycle. There were methodological problems and a single year record in the case of the Harris study, and in this one below average values extended from October right through spring in the second year. One might conjecture that the DON pool could be depleted by bacterial activity when phytoplankton production was limited by light because algae are such an important source of DON. Harris (1959) suggested the connection between DON and phytoplankton because DON increased in June along with a bloom of dinoflagellates. In the summer, or whenever DIN is depleted, DON becomes singularly important as a source for DIN regenerated by microbes. The present view emphasizes the importance of very rapid rates of nitrogen release and uptake in the water column, which produce a tight coupling between heterotrophic and autotrophic processes, while there is often virtually no detectable change in the pool of free dissolved nitrogen as a whole (Nixon & Pilson, 1983). We suspect that the higher DON observed in summer helps maintain primary production rates by increasing the rate of regeneration by the microbes.

The eastern station at Hammonasset had generally fewer copepods than has been observed in other sudies; however, stocks at Milford and Stamford were about as expected, except for unexpectedly small summer populations (Deevey, 1956; Peterson, 1986; Dam, 1989). Deevey's copepod abundance data are spatially averaged, and both Dam and Peterson worked west of Hammonasset, so the lower numbers of copepods at Hammonasset may not be unusual. Copepod biomass in the spring was dominated by *Temora longicornis*, and the peak biomass estimates for that species at Milford and Stamford were not significantly different from Dam's (1989). The species composition was in most ways indistinguishable from Peterson (1986) and Deevey (1956). However, both Deevey and Peterson found *Oithona* sp. more important during the summer and fall seasons than we did. The 202-μm net may not have retained many individuals of *Oithona* sp., based on the small narrow shape of the few individuals that did appear in our samples.

The typical lack of the summer copepod peaks that were seen by Deevey (1956) and Peterson (1986) suggests that the summer food-web may have changed somehow. Perhaps ctenophores are more abundant (our ctenophore debris observations would support this notion), or perhaps a shift has occurred away from copepods. However, it would be prudent to remember that there are inshore-offshore differences, and considerable year-to-year variability in LIS, before coming to any conclusions (although this study does represent a multi-year data set). The 202-μm net counts ranged from 183 to 59542 ind m^{-3}, Deevey's (1956) 158-

μm net counts ranged from about 5000 to 150000 ind m^{-3}, and her 363-μm net counts ranged from about 200 to 30000 ind m^{-3}. A 158-μm mesh size retains more individuals than a 202-μm mesh, including small juveniles, whereas a 363-μm mesh retains fewer individuals than a 202-μm mesh. The fact that the counts were generally bracketed above and below by counts Deevey made with nets above and below in mesh size suggests that although changes may have occurred since 1956, they are not drastic changes.

Our 3 year larval fish work indicated that fish diversity was down as compared to S. Richard's and G. Riley's 1950s data. Overall, abundance was similar across our 3 east – west stations. When differences were encountered, they indicated higher levels at our western station. Also, over the course of 2 years of oxygen measurements at our 40-foot depth stations, hypoxic conditions were only encountered once. Given the above findings, larval fish should not experience food limitations nor severe oxygen stress, at least in near-shore waters. Thus, if juvenile and adult fish stocks are down, one must look elsewhere for a cause. Our species compositional work presented above (see results section on larval fish) indicates that two major factors may be overfishing and predation pressure (e.g., cormorants). If larval fish fail to capitalize on the high copepod biomass, ctenophores and other gelatinous zooplankton may be the beneficieries, i.e. we may be enhancing economically uninteresting gelatinous zooplankton at the expense of fish (Grieve-Parsons hypothesis, 1977). Indeed, anecdotal information and our ctenophore debris observations suggest that ctenophores are enhanced in the waters of the western Long Island Sound.

Conclusions and summary

Yearly and multi-year average concentrations of dissolved inorganic nutrient levels indicate similar (to only slightly higher in the west) nutrient concentrations across an east to west LIS gradient.

The relative proportioning among chemical species of nutrients often differs from west to east both contemporaneously as well as temporally, with NH_4 and DON at times more prevalent in the west (particularly in bottom waters).

The excess loading of nitrogen (and other nutrients) into the Sound appear to be converted to elevated biomass in the west (as evidenced by observed chlorophyll, phytoplankton and zooplankton concentration gradients).

Size-fractionated chlorophyll data indicate little east to west differences in the 10–20 μm size fraction while pointing to large differences in the <10 and >20 μm fractions which are both higher in the west.

Occurring along with the enhanced phytoplankton biomass is slightly enhanced bacterial densities and growth rates. The densities show interesting seasonal bacterial cycles and appear to be related not to total chlorophyll levels but to densities of the photosynthetic nanoplankton (PNANs).

Heterotrophic nanoplankton densities (HNANs) also are higher in the west and also appear to at times influence bacterial densities.

Species composition of phytoplankton routinely differ among west to east stations. These species compositional shifts appear to be related to N/P and N/Si ratios as well as to ratios among nitrogen chemical species.

Dissolved inorganic N/P ratios are routinely low among all stations with the west exhibiting lower levels than the east. However, total dissolved nitrogen /P ratios (which includes DON) are similar among stations and typically above the Redfield ratio of 16:1.

Associated with enhancement of bacterial, HNAN and <10 μm chlorophyll is significant enhancement of ciliate species diversity.

Microcrustacean (copepod) biomass also is extremely enhanced in the west vs. the east indicating that while stimulating the microbial loop, eutrophication is also enhancing secondary production preferred by larval fish, comb jellies and other jellyfish.

Copepod biomass as well as fecal pellet production likely is a significant contributor to hypoxia in Long Island Sound.

Our 3 year larval fish work indicated that fish diversity was down as compared to S. Richard's & G. Riley's 1950s data. Overall, abundance was similar across our 3 east to west stations. When differences were encountered, they indicated higher levels at our western station.

Over the course of 2 years of oxygen measurements, at our 40-foot depth stations, hypoxic conditions were only encountered one time. Given the above conditions larval fish should not experience food limitations nor severe O_2 stress, at least in near shore waters. Thus if juvenile and adult stocks are down one must look elsewhere for a cause.

If larval fish fail to capitalize on the high copepod biomass, ctenophore and other gelatinous zooplankton may be the beneficiaries, i.e. we may be enhancing economically uninteresting gelatinous zooplankton at the expense of fish (Grieve-Parsons hypothesis). Indeed, anecdotal information suggests that ctenophores are enhanced in the waters of the western Long Island Sound.

Dual enhancement

Results from the completed study indicate that both the microbial loop as well as the copepod sector of the food web are enhanced west of Hammonasset. One implication of this 'dual enhancement' is that, in this instance, increased microbial activity does not take food away from the copepods. Since copepods are omnivorous, the increased numbers of protozoa may in fact have contributed to the increase in copepod standing stocks between Hammonasset and Milford.

New conventional wisdom

Excess nitrogen stimulates microbial loop and net phytoplankton biomass/production which in turn stimulates microcrustacean biomass/production, and fecal pellet release, which likely significantly fuel hypoxia, and whose ultimate fate is at this time unclear, but may very well be both gelatinous zooplankton as well as the sediments of Long Island Sound.

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The feeding strategy of the dunlin (*Calidris alpina* L.) in artificial and non-artificial habitats at Ria de Aveiro, Portugal

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Key words: waders, dunlin, natural mudflats, salt pans, high tide feeding, Ria de Aveiro

Abstract

Usually waders (Aves, Charadrii) feed in the inter-tidal zone over the low tide period and roost over high tide. But occasionally when their energetic needs increase, due to adverse weather conditions or to a need to store fat to fuel migration, the birds also feed during high tide, sometimes in agricultural land. At the Ria de Aveiro, about 44% (corresponding to 4500 birds) of the wintering dunlin (*Calidris alpina*) use artificial salt pan habitats throughout the winter whilst the others utilize natural intertidal habitats, mainly mudflats. The dunlin that feed intertidally in natural areas, such as the Canal de Mira, do not feed over high tide. In contrast, the birds that use the salt pan area feed both during low tide, in abandoned salt pans whose walls are breached and thus allow access to the tide, and high tide, in active salt pans, and keep on doing so throughout the winter. The energetic needs of both groups of birds are thought to be similar. However, the benthos of the abandoned salt pans seem to be different, and poorer in energetic terms, from those of the natural mudflats. It is argued that dunlin wintering in the salt pan area must compensate for this by feeding over high tide in active salt pans. Rather than just being a means of obtaining more energy at certain times of high energy demand, it would seem that feeding in active salt pans over high tide should be considered as a normal component of the feeding strategy of these birds, a strategy that is different from those wintering in the natural mudflats. These findings highlight the importance of the salt exploitation industry to waders and also show that artificial habitats can support large numbers of wintering waders.

Introduction

Salt pans are artificial habitats that usually support large numbers of waders, as was already shown by several authors (e.g. Rufino et al., 1984; Perez-Hurtado & Hortas, 1991, 1993a, b; Santos, 1997). In some areas, salt pans seem to be more important than other more natural habitats, to the extent that some waders concentrate their activities there, irrespective of the state of the tide (Velasquez & Hockey, 1992).

Salt exploitation has been carried out at the Ria de Aveiro for many centuries and, by the 15th century, salt pans occupied a large part of this wetland. Nowadays, large numbers of salt pans are being abandoned, following the decline in the salt exploitation industry (Luís, 1999). The walls of most abandoned salt pans erode and, as they are not maintained, they are eventually breached, allowing the tides to go in and out. For waders, this has the effect of turning the former salt pans into habitats that may probably be best defined as artificially-created 'natural' inter-tidal flats. In contrast, active salt pans are not subjected to tides, the water levels inside are usually kept low and they can be used by waders as roosts or feeding areas at any stage of the tide.

During preliminary studies at the Ria de Aveiro, we observed that dunlin (*Calidris alpina*) wintering on the salt pan area fed both over low tide in abandoned salt pans and over high tide in active salt pans and kept on doing so throughout the winter. The walls of abandoned salt pans have been breached and so the tide can come in and out and new mudflats thereby develop on which the birds can feed over the low tide exposure period. In contrast, the walls of the active salt pans are, of course, maintained, and the water levels remain more-or-less constant throughout the tidal cycle.

In contrast to the salt pan area, the dunlin wintering on nearby natural intertidal habitats fed only on the intertidal mudflats when these were exposed over low tide, and thus occupied their feeding grounds for a much shorter period each day. As the energetic needs of both groups of birds were expected to be similar (Luís, 1999), it appeared that dunlin feeding on the artificial salt pans required a much longer period of time to obtain their requirements than those that fed on the natural mudflats. This, in turn, implied that the quality of the salt pans as feeding areas for dunlin was not as good as the quality of the natural mudflat areas.

Thus, it seems that the dunlin wintering on the salt pan area may be adopting a different feeding strategy to those wintering on the natural mudflats area and this difference may be related to differences in the benthic fauna and/or to the length of the exposure period. Nonetheless, at present, the salt pan area (that occupies 13% of the total area of the lagoon) supports about 44% (corresponding to 4500 birds) of the dunlin wintering in Ria de Aveiro. Therefore, understanding the way in which waders such as the dunlin use the salt pans and survive winter could have important implications for the management of this area in particular as well as to the planning and restoration of similar, and other types of artificial habitats, elsewhere.

In order to understand better the role played by the salt pans of the Ria de Aveiro in the winter feeding ecology of the dunlin, we investigated the amount of time spent feeding by birds of both groups on active and abandoned salt pans, and on non-artificial natural mudflats. In particular, we wanted to understand how artificial feeding areas, that at first sight seem to be of poor quality compared with natural mudflats, nonetheless supported almost half the wintering dunlin population. The results and interpretation of this work are presented here.

Study area and methods

The Ria de Aveiro is a coastal lagoon, located in Portugal, on the West coast of the mainland (40° 50′ N). The lagoon has three main arms that radiate from the mouth (Canal de Ovar, Canal de Ílhavo and Canal de Mira) and a large central area occupied mainly by salt pans and some fishponds (Fig. 1).

The selected study areas were the natural mudflats of the Canal de Mira and the central salt pan area, where both active and abandoned salt pans were studied. Preparatory studies, using radio-tracking, counts of dye-marked dunlin and counts of the total number of dunlin in each area did not reveal the existence of movements between the two study areas (Luís, 1999). Most of the work was carried out during the 1995–1996 winter.

Observations were made through a telescope (20–60×77) from a hide in the salt pan area and from a vehicle in the Canal de Mira. The high and low tide observation periods started when the first birds arrived in the study site and ended only when the last birds had left. The number of observation periods is represented by '*n*' in the legends for the figures. Birds were counted every 30 min, the numbers feeding and not feeding being counted separately.

Preliminary visits had shown that, in both the Canal de Mira and salt pan area, the low tide feeding period started as soon as some small areas of sediment became exposed. As the tide receded, most birds then moved on to other feeding grounds. For this reason, the results for the feeding grounds used by the birds at the beginning and the end of the exposure period are shown separately from those for the grounds used during the middle part of that period.

Results

Salt pans

The observations made during the high tide period in the active salt pans showed that most dunlin (65.7%±10.71) started to feed upon arrival from their low tide feeding grounds (Fig. 2). The proportion of birds feeding decreased, more or less steadily, to 41.4% (±7.04) at the time of high tide, when the average number of birds present was very high (1277.1±271.39). Dunlin started to leave the area about 1 h after high tide. But by 2.5 h past high tide, when most birds had already left, 37.0%(±8.12) of the remaining dunlin were still feeding. The feeding activity stopped shortly after this but a small number of birds did not leave for their low tide feeding grounds until 4 h past high tide. Taking the 5.5 h period from 3 h before to 2.5 h after high tide as a whole, the average proportion of birds feeding was 44.0% (±2.46).

Birds moved to the low tide feeding grounds, the abandoned salt pans, as the tide receded as soon as the first areas of sediment became visible there (Fig. 3). However, in this initial stage (3.5 h before low tide), more than 50% of the dunlin were not feeding.

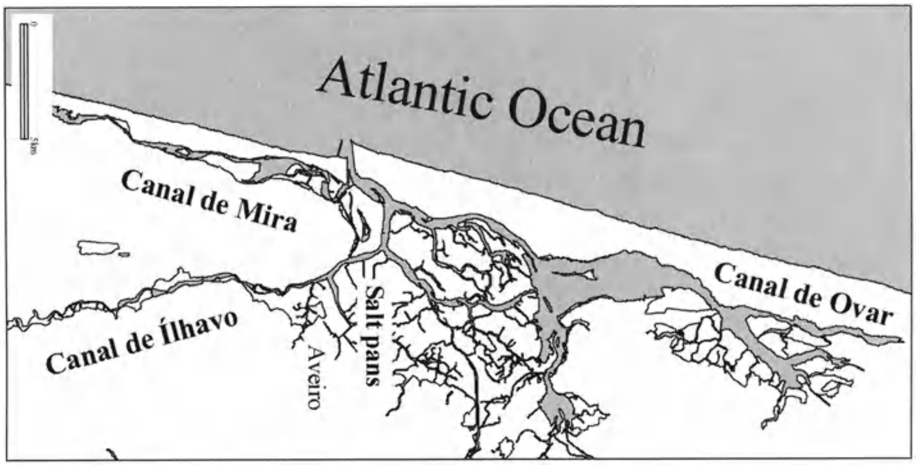

Figure 1. The Ria de Aveiro.

As can be seen from Figure 3, the proportion of birds feeding increased rapidly to 92.9% (±7.15) by 3 h before low tide. Apart for two slight decreases just before and just after low tide, the proportion feeding was relatively constant and remained high until the end of the exposure period. Over the exposure period as a whole, the average was 87.1%±3.16.

The number of birds present also increased through the initial stages of the exposure period, to reach a maximum of 2296.7 (±340.65) – corresponding to 51% of the total number of birds in the salt pan area – 1 h after the sediments became exposed. Subsequently, the numbers declined towards low tide, as birds moved to more recently exposed areas downshore. Most dunlin started to feed as soon as they arrived at these downshore areas (Fig. 4). The proportion of birds feeding in the downshore areas was remarkably high (93.5%±2.72) and stable throughout the exposure period.

Figures 2 and 3 also show that, the active salt pans, used over high tide, and the abandoned salt pans, used over low tide, were each exploited by dunlin for a maximum period of 7.5 h. Therefore, in total, the two kinds of salt pans were used for 15 h over a single tidal cycle. As a tidal cycle lasts for about 12.5 h, there was clearly an overlap in the periods for which the two kinds of salt pans were exploited by dunlin. This was due to the fact that some birds arrived earlier and/or left the low tide feeding grounds later than others, while other birds stayed for longer in the active salt pans. In order to calculate the amount of time spent feeding by an average bird in both the active and abandoned salt pans, while dealing with this problem of overlap in feeding periods, we assume in the calculations referred to in the 'Discussion' that the high tide and low tide periods lasted for 5.5 (3 h before to 2.5 h after high tide) and 7 h (between 3.5 h before and after low tide), respectively.

Natural mudflats (Canal de Mira)

In the natural mudflats, most birds started to feed as soon as they arrived on the feeding grounds. At the beginning of the exposure period, 3.5 h before low tide, 1560.0 (±5.77) dunlin – corresponding to 84% of the total number of birds using the Canal de Mira – were actively feeding on the partially exposed feeding ground (Fig. 5).

Subsequently, they then dispersed to other more recently exposed areas downshore as a consequence of which, the number of birds dropped to 326.7 (±12.02) only half an hour later. However, and in contrast to their behaviour in the salt pan area, the birds immediately resumed feeding as soon as they arrived on the newly exposed grounds (Fig. 6). The proportion of birds feeding was always at (or very near) the maximum of 100% until the end of the exposure period, except for two short periods, at low tide and 1 h after, when some birds would suddenly stop feeding, thus lowering the percentages values towards 80% (Fig. 6).

As the tide advanced and the downshore areas became flooded, dunlin returned to the upshore areas they had used at the beginning of the exposure period where, again, they immediately resumed feeding until the advancing tide drove them from the mudflats altogether (Fig. 5).

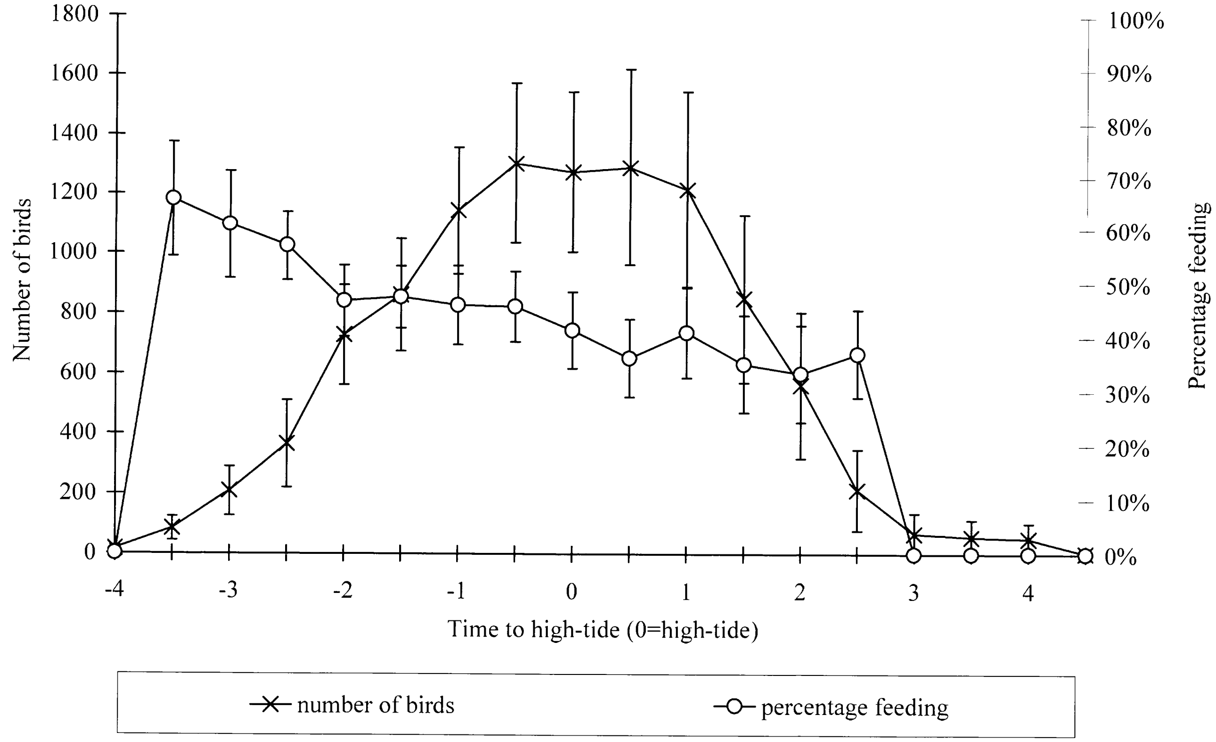

Figure 2. Salt pan area (active salt pans). Average number of birds present (±1 se) and the percentage feeding (±1 se) during high tide in three salt pans (n=7).

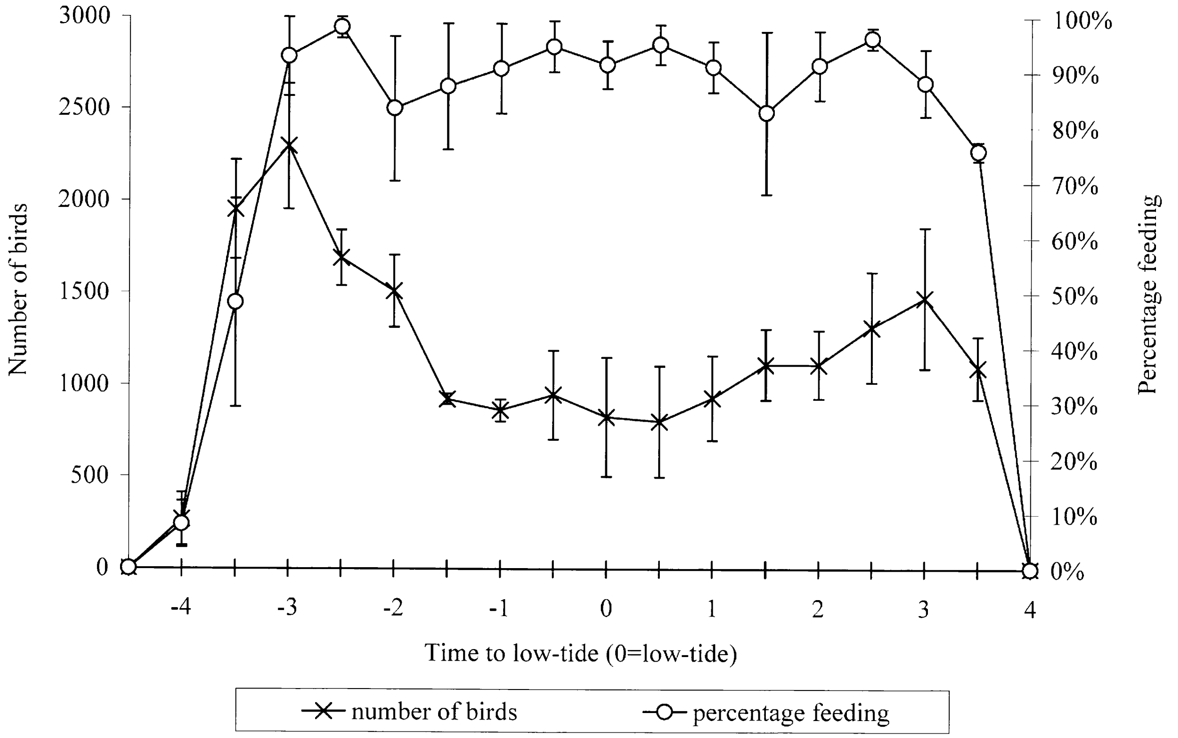

Figure 3. Salt pan area (abandoned salt pans). Average number of birds present (±1 se) and the percentage feeding (±1 se) in an abandoned salt pan used by 51% of the dunlin as a low tide feeding ground, at the beginning and end of the exposure period (n=3).

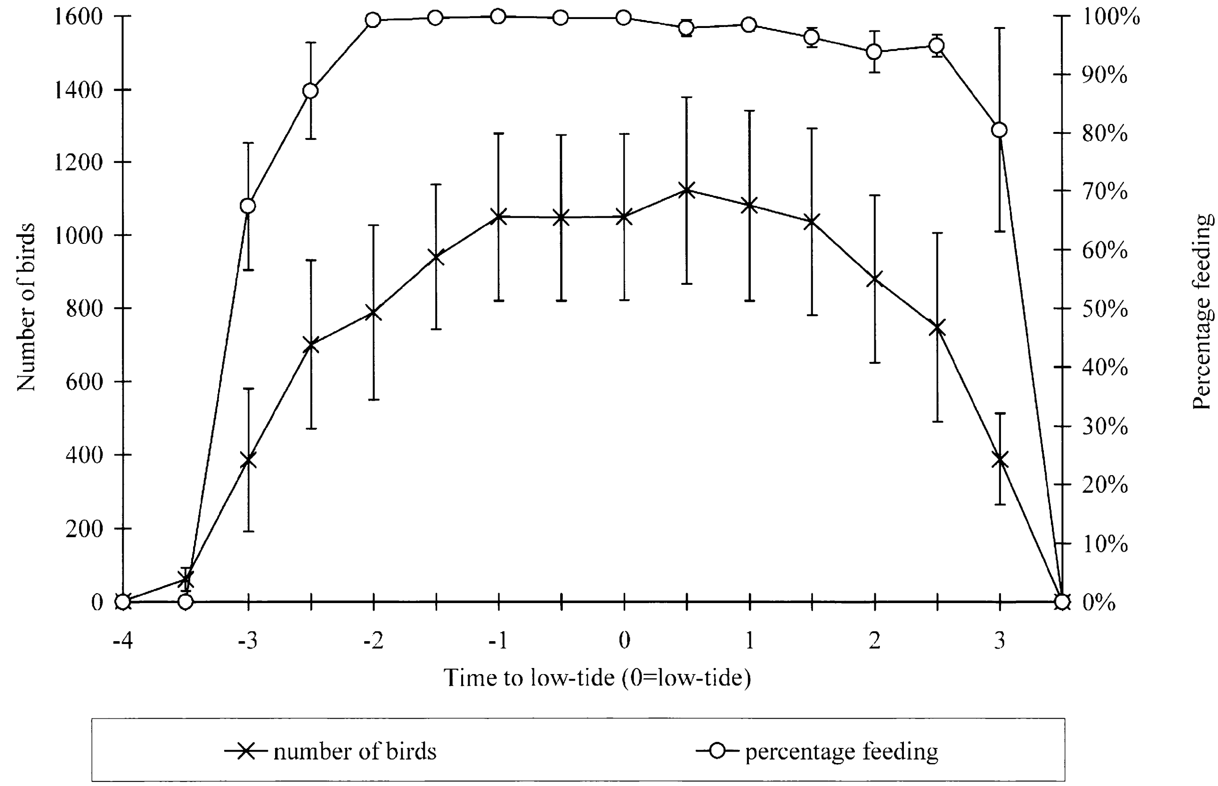

Figure 4. Salt pan area (abandoned salt pans). Average number of birds present (±1 se) and the percentage feeding (±1 se) in 3 abandoned salt pans used by dunlin as their low tide feeding grounds over the middle of the exposure period (*n*=5).

Discussion

Main findings

The duration of the exposure period is similar in the natural mudflats and salt pans. Yet the dunlin wintering in the Canal de Mira fulfil their energetic demands during the low tide period only, whereas the birds wintering in the salt pans get part of the energy they need from active salt pans, where they feed over high tide.

At first sight, this might suggest that the birds feeding on natural mudflats obtain their energy requirements in less time each tidal cycle than do the birds in the salt pans. On the other hand, feeding activity patterns over low tide are somewhat different in the two areas. Whereas the birds in the natural mudflats all start feeding immediately upon arrival on the receding tide and keep on doing so throughout the exposure period, stopping only occasionally, some of the birds that feed in the abandoned salt pans do not start feeding until after the tide has receded much further. Furthermore, the proportion of birds feeding in the abandoned salt pans seems to be lower than in the natural mudflats, and the pauses in feeding are more asynchronous.

The birds that use the natural mudflats compress all their feeding into the exposure period and do not use feeding grounds over high water, even though they could easily fly to the active salt pans and feed there. In contrast, the dunlin in the salt pan area feed in the abandoned salt pans over low tide and also over high tide in the still active pans. The results may therefore indicate that the dunlin wintering in the salt pan area seem to adopt a feeding strategy that is adapted to resource availability and, probably, to the lower quality of the low tide feeding grounds. This possibility is now discussed in more detail.

Natural mudflats (Canal de Mira)

As can be seen from Figure 5, the feeding period on the natural mudflats lasts for about 7 h. The average proportion of feeding birds over the whole 7 h period can be calculated from the data in Figures 5 and 6 by combining the percentage values for the period between 2.5 h before and after low tide (Fig. 6) with the percentage values for the −3.5 to −3 h and the 3

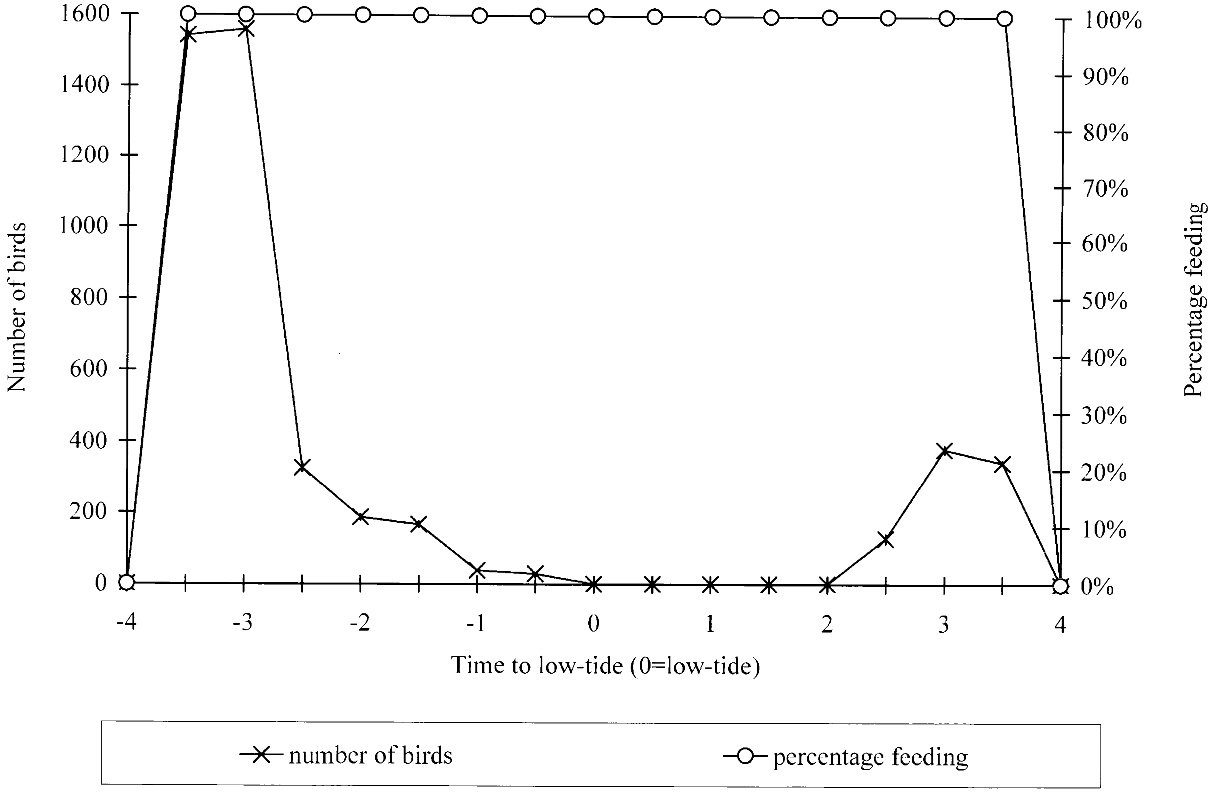

Figure 5. Natural mudflats. Average number of birds present (±1 se) and the percentage feeding (±1 se) on a mudflat used by 84% of the dunlin at the beginning and end of the exposure period (*n*=3).

to 3.5 h (Fig. 5). Calculated this way, the proportion of birds that fed over for the 7 h period as a whole was 95.9% (±1.32).

Following Goss-Custard (1969), the percentage of birds feeding measures the proportion of the available feeding time that the average bird spends in feeding activities. Thus, on the natural mudflats area, the average bird fed for 6.7 (±0.09) hours per tidal cycle.

Salt pan area

The same calculation can be made for the birds feeding over low tide in the abandoned salt pans. This was done by combining the percentage values for the 5 h period (−2.5 to 2.5 h before and after low tide) in Figure 4 with those for the −3.5 to −3 h and the 3 to 3.5 h before and after low tide in Figure 3. The average proportion of birds feeding over the 7 h low tide exposure period was 87.5% (±1.01). Thus, the average bird would have fed for 6.1 (±0.14) h over the low tide exposure period in the abandoned salt pans.

However, in the salt pan area birds also fed over high tide in the active salt pans. Therefore, the time spent feeding then must also be considered when calculating the total amount of time spent feeding per tidal cycle. According to the data, 44.0% (±2.46) of the dunlin fed over the 5.5 h high tide period and, consequently, the average bird fed for 2.4 (±0.35) h. However, active salt pans were sometimes flooded during periods of heavy rainfalls, and this prevented birds from feeding there. If it is assumed that the salt pans were flooded for, at the most, one third of the winter, the average amount of time spent feeding over high tide by the average bird is reduced to 1.6 h. Hence, in the salt pan area, the average bird feeds for 7.7 h per tidal cycle, which is 1 h more than the birds on the natural mudflats area fed.

Interpretation

Comparison between the quality of feeding conditions in the salt pans and natural mudflats

Radio-tracking studies have shown that the distances flown per tidal cycle by the dunlin wintering in the salt pan area and the natural mudflats area are not significantly different, so that the energetic require-

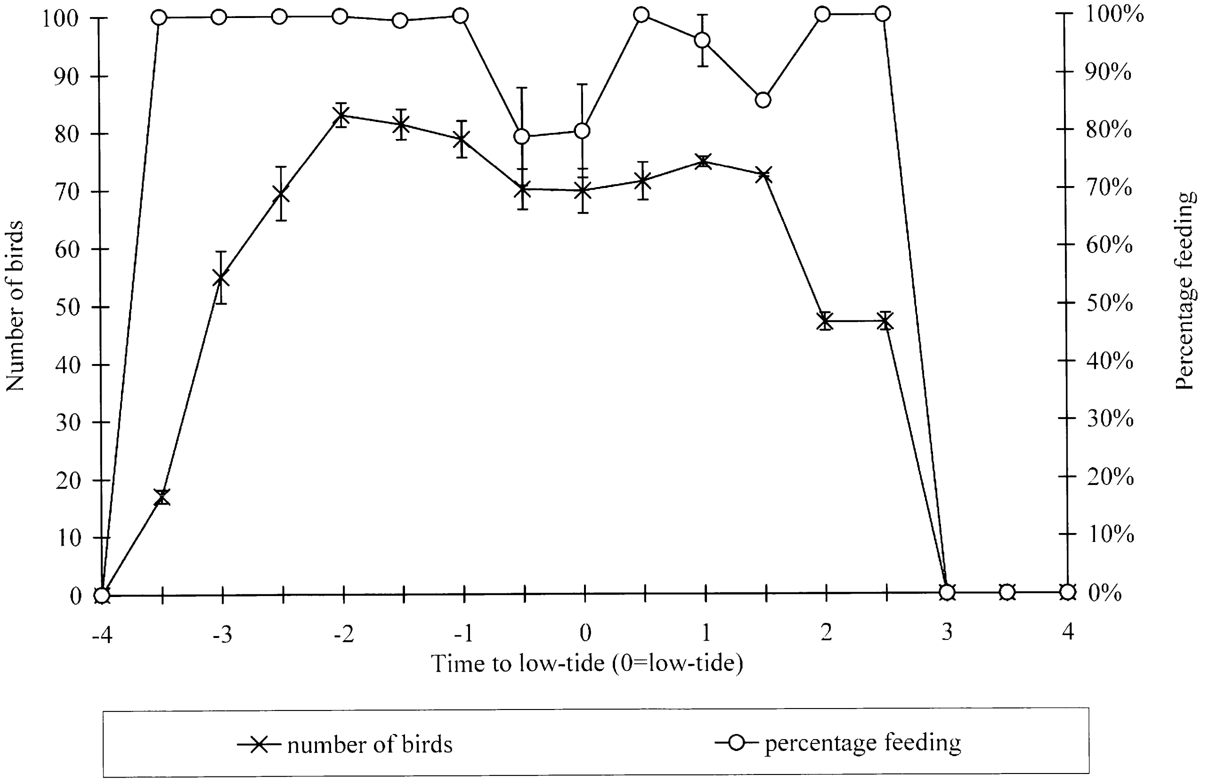

Figure 6. Natural mudflats. Average number of birds present (±1 se) and the percentage feeding (±1 se) on a mudflat used by dunlin over the middle of the exposure period (n=3).

ments of both groups of birds would be expected to be similar. Furthermore, no differences were found in the body condition, weight, age or sex-ratio of both groups of birds (Luís, 1999). So, in order to fulfil the same energetic needs, the dunlin wintering in the salt pan area feed for 1 h longer than do those wintering in the natural mudflats area. This might support our initial hypothesis that feeding conditions were generally better in the natural mudflats than in the salt pan area.

On the other hand, over the main low water feeding period, the birds wintering on the salt pan area fed for less time than did the birds on the natural mudflats (Mann–Witney, U=15 p<0,05; n_1=3, n_2=5). This might suggest, therefore, that the feeding conditions in the abandoned salt pans, at least, were actually better than those in the natural mudflats. Alternatively, it could simply mean that the two groups of birds do not use the same feeding strategy.

According to rate-maximising foraging theory, predators will tend to maximise their ingestion rate (biomass ingested per unit time) while actively foraging. However, as has been shown for the oystercatcher *Haematopus ostralegus* (Kersten & Visser, 1996), the ingestion rate of feeding birds may often be higher than the gut processing rate (biomass digested per unit time). When the ingestion rate exceeds the digestion rate, a bird will have to stop feeding as soon as the oesophagus is full to allow for digestion (Zwarts & Dirksen, 1990). This could explain the decrease in the percentage of birds feeding that was observed around low tide in all feeding grounds. This could also explain the lesser amount of time spent feeding during low tide by the dunlin wintering at the salt pans. As the salt pan area birds fed during high tide and arrived on the low tide feeding grounds with the gut at least partially full they might eventually have to stop feeding to allow for digestion at some stage during the beginning of the emersion period. Moreover, this could also explain the differences between the natural mudflats and the abandoned salt pans in the pattern of variation in the proportion of birds feeding. Thus, while the greater amount of time spent feeding by the dunlin in the salt pans overall does suggest that these areas provide poorer feeding conditions than the natural mudflats, there are some grounds for being cautious in accepting this interpretation and other comparisons are needed.

A more direct comparison of the feeding conditions could be made by comparing the food supply present in the two areas. In her study of the benthic fauna of abandoned salt pans, A. Ravara (unpublished results) showed that species diversity was low but the densities were high compared with the natural mudflats. She also stressed that the benthic organisms in the salt pans were unusually small in size. In contrast, more than 50% of the species found by Moreira (1988), Moreira et al. (1993) and Rocha (1995) in the natural mudflats of Canal de Mira were represented by individuals whose size was, usually, over 1 cm. Since prey size is an important determinant of ingestion rate in waders (Goss-Custard 1977; Zwarts et al., 1996), these findings might further suggest that the abandoned salt pans did indeed provide poorer feeding conditions than the natural mudflats.

A study of the birds' diets in the Ria de Aveiro also supports this idea: 29.2% of the droppings collected during low tide in abandoned salt pans had no identifiable remains, as opposed to 0% in the ones collected on the natural mudflats. This suggests that the birds feeding in the salt pans captured small and/or soft bodied prey more often than did the birds on the natural mudflats (Luís, 1999). This implies that the average size of the benthic organisms taken from the natural mudflats by dunlin would then be higher than those in the abandoned salt pans, leading to higher ingestion rates.

Overall, we conclude that our preliminary evidence does conform with the hypothesis that the feeding conditions provided by the natural mudflats for dunlin are superior to those provided by the abandoned salt pans.

Pattern of foraging through the tidal cycle in the salt pans and the natural mudflats

If our conclusions on the relative rates of ingestion in the two habitats are indeed correct, we can attempt to interpret the differences between the areas in the patterning of feeding activity through the tidal cycle. According to Zwarts et al. (1996), an oystercatcher that leaves the feeding grounds with the digestive tract completely full will take about 5 h to completely digest the food. Five hours is more or less the duration of the high tide period at Ria de Aveiro but, as dunlin are much smaller than oystercatchers, their gut would be expected to empty much more quickly than those of oystercatchers. Therefore, at the beginning of the emersion period, the digestive tract of the birds that feed on the natural mudflats will presumably be completely empty. They should therefore be able to feed continuously for a number of hours, depending on the ingestion rate.

In contrast, high tide feeding occurs in the salt pan area, in active salt pans. Thus, on the assumption that ingestion rates are higher than digestion rates, the birds whose digestive tract is at least partially full upon arrival on the low tide feeding grounds will stop feeding at the very beginning of the exposure period, or shortly afterwards. This may account for the low proportion of feeding birds at the beginning of the exposure period as well as for the small decrease observed 2 h before low tide (Fig. 3). As a result, the pauses to allow digestion might be expected to be more asynchronous in the abandoned salt pans than in the natural mudflats, where all birds begin feeding at the same time at the end of the high tide period. And for the same reason, the average proportion of birds feeding over the low tide exposure period as a whole would be lower, when compared to those at the natural mudflats.

Conclusions

These results point out that high tide feeding in active salt pans should not be considered just as a means to obtain supplementary energy but, more exactly, as an essential part of the feeding strategy of these birds, in which active salt pans play a fundamental role. According to this interpretation, these two groups of birds wintering at Ria de Aveiro adopt feeding strategies that are different and adapted to the specific characteristics of the areas where they settle.

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Short-term variations in the physiological state of phytoplankton in a shallow temperate estuary

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Abstract

Short-term changes in the photosynthetic carbon metabolism and physiological state of phytoplankton were studied over a summer fortnight-long period in the Urdaibai estuary (Bay of Biscay) and related to observed environmental patterns. Day-to-day variability in the hydrographical and biological features of the estuary during the study period was due to changes in meteorological and tidal conditions. Phytoplankton biomass and primary production increased with the improvement of weather, i.e., light conditions, during neap tides. Thus a mixed bloom of cryptophyceans, *Euglena* sp., and the dinoflagellate *Peridinium foliaceum* developed in the middle and upper estuary. Photosynthetic responses of phytoplankton were related to the time-scale of changes in light regime. Allocation of photosynthate to major macromolecular classes (LMWM, lipid, polysaccharide, and protein), like phytoplankton biomass and primary production, showed strong spatio-temporal variability. High carbon fixation into low molecular weight metabolites was associated with growth limitation by low light. The relative incorporation of photosynthetic carbon into proteins increased at the beginning of the phytoplankton bloom but overall, it was rather constant. However, carbon allocation into storage products such us lipid or polysaccharide increased when carbon and energy produced under optimal growth conditions exceeded what could be assimilated into protein. These patterns are explained by both spatio-temporal changes in the environmental conditions and species-specific differences. In general, daily variability appeared to be more important than diurnal periodicity in the physiological responses of phytoplankton. Results from this study show that phytoplankton photosynthesis and carbon metabolism are simultaneously affected by biotic and abiotic factors, although short-term light fluctuations may have a major influence on the physiological state of phytoplankton in the Urdaibai estuary.

Introduction

The ultimate goal of the determination of phytoplankton physiological state is to relate the growth rate to intrinsic algal properties. If such a functional relationship is known, one can on basis of these properties decide whether an algal population grows at the maximum rate or not, and in the last case find how much the rate is reduced and by which factor the reduction is caused. Thus, the concept of physiological state implies nothing more than an indirect estimate of the production of algal matter with the implicit knowledge of growth-regulating factors (Sakshaug, 1980).

A wide range of different algal properties have been used to estimate the physiological state of phytoplankton. Among them, cell activities, biochemical composition, and morphological characteristics have proved to be useful physiological indicators, even when species specific variability is considered (Zevenboom, 1986). The most commonly investigated physiological processes are photosynthesis, carbon and nitrogen metabolism, nutrient uptake, and enzymatic activities (Platt, 1981); biochemical studies generally include determinations of the elemental, macromolecular and pigment composition (Madariaga & Joint, 1992). Visual indicators, however, are strictly qualitative and time-consuming, and consequently they have not been applied widely in research on natural populations. Both physiological and biochemical measurements are good indicators of different time-scale processes. As discussed by Morris (1981), physiological measurements of cellular activity must be interpreted with respect to the immediate cell environment, and they will indicate potential future changes in cell biochemical composition. In contrast, the actual biochemical composition reflects the physiological history of the cell.

Phytoplankton have the capacity to rapidly adjust their photosynthetic metabolism in response to changes in environmental conditions. This physiological adaptation involves a complex series of interacting processes occurring at different time scales, which define the physiological state. Knowledge of these processes is crucial to understanding the phytoplankton ecophysiology in any aquatic ecosystem. In particular, temperate estuaries are subjected to a high short-term variability depending on freshwater inflow (freshets), tides (neap-spring), wind (storms), and other episodic events (Cloern & Nichols, 1985). In many cases, these processes drastically modify the estuarine habitat properties, which lead to pronounced fluctuations in the composition and dynamics of natural phytoplankton communities. Although it is well known that the physiological characteristics of phytoplankton are variable over a wide range of spatio-temporal scales (Harris, 1980), the role of short-term processes in estuarine phytoplankton ecophysiology, i.e., those occurring at time-scales which approximate cell division times, has historically received little attention (Litaker et al., 1987, 1993).

In this context, physiological parameters such us the different incorporation patterns of photosynthetic carbon into macromolecules can be used to monitor phytoplankton natural populations, because such measurements provide significant information about the physiological state of algae which is not obtained by estimations of total photosynthetic carbon fixation. While macromolecular synthesis patterns are controlled to a large extent by the immediate cell environment, such as the prevailing light, nutrient and temperature conditions (Hitchcock, 1983), the latter depend on relatively slower alterations in levels of photosynthetic pigments, electron transport proteins, or enzymes (Konopka, 1982). As stated by Morris (1981), the study of photosynthate partitioning reveals the immediate physiological responses of the algae to their growth conditions on short-time scales.

The study of such patterns is critical because in many cases there is a poor relationship between photosynthetic carbon uptake and cell division (Smith & Morris, 1980). High rates of photosynthesis may, for example, represent an accumulation of reserve products rather than cell growth (Hama et al., 1988). However, there is a significant correlation between the photosyntetic protein production and phytoplankton growth (Morris, 1981; Taguchi & Laws, 1987; Hama et al., 1988; Fernández et al., 1994). These variations in the incorporation patterns of photosynthetic carbon into macromolecules depending on the physiological state of phytoplankton, affect their quality as a food source, and ultimately the whole food chain dynamics within the ecosystem.

To date, only a few studies of estuaries and coastal zones have provided a comprehensive examination of short-term temporal and spatial variability in phytoplankton photosynthesis, and attempted to delineate the factors causing this variability (Côte & Platt, 1983; Shephton & Harris, 1984; Madariaga & Orive, 1989; Madariaga et al., 1989; García-Soto et al., 1990; González-Morales et al., 1993; Madariaga, 1995; Delgadillo-Hinojosa et al., 1997). However, no attempt has been made yet to determine short-term metabolic patterns of phytoplankton along the strong spatial and temporal gradients in environmental conditions that occur in estuaries.

In this work, short-term variability in the physiological state of phytoplankton was studied on a daily time scale in the Urdaibai estuary. The study area is characterized by a high degree of spatial and temporal variability under unstable atmospheric conditions (Madariaga & Orive, 1989). During spring and summer, intense phytoplankton blooms commonly develop in the upper reaches of the estuary, where nutrient concentrations are high due to anthropogenic inputs (Madariaga et al., 1989). The sampling strategy was designed in such a way that the physical and chemical causes of phytoplankton short-term variability during the occurrence of typical summer blooms in this estuary could be studied. Thus, a complete spring-neap tidal cycle was covered during a fortnight-long study period, characterized by variable meteorological conditions. Diurnal variations in phytoplankton photosynthetic properties were also investigated, as this time scale of variation is superimposed on the day-to-day variability.

The final aim of this work was to describe and interpret the photosynthetic carbon metabolism and physiological state of estuarine phytoplankton in relation to observed short-term environmental patterns in a highly variable ecosystem such as the Urdaibai estuary.

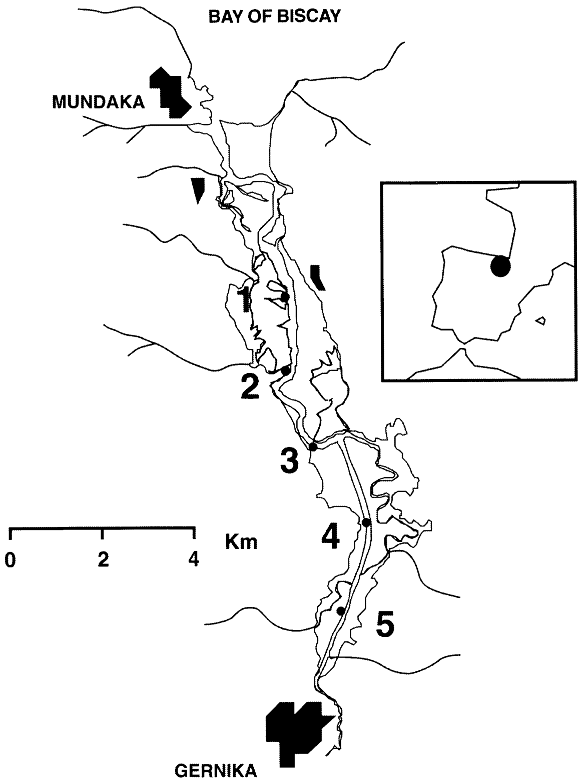

Figure 1. Study area and sampling sites.

Materials and methods

The Urdaibai estuary (also known as Gernika estuary) is a mesotidal temperate estuary located in the Basque Country (43°22′ N, 2°40′ W), North of Spain (Fig. 1). It is rather shallow (mean 2.6 m), and extends approximately 12.5 km from its mouth to the city of Gernika, with a steep slope (mean 0.201 m km^{-1}). It is bordered by relatively extensive tidal flats along its lower reaches and by salt-marshes in the middle. These geomorphological features determine the estuarine ecosystem, as it can vary considerably in its volume and flushing rates, which strongly affects its biological and chemical properties (Madariaga, 1995; Madariaga & Ruiz, 1988; Madariaga & Orive, 1989; Madariaga et al., 1989, 1992, 1994).

The study was carried out from 3 to 15 July 1993. Daily surveys were conducted at five permanent sampling stations distributed along the longitudinal axis between the outer estuary and the middle of an artificial channel which joins the river Oka near Gernika (Fig. 1). This point normally represents the upper limit of sea water penetration. Water samples were taken from the subsurface (0.5 m), always at high tide, with a 5-l opaque Van Dorn bottle. Only subsurface waters were considered in this study because bottom samples mainly reflect sediment resuspension processes (Madariaga & Orive, 1989). At each station vertical temperature profiles of salinity, temperature, pH and oxygen content of the water were determined using WTW sensors that were calibrated just before each survey. Profiles of photosynthetically active radiation (PAR) were also obtained from surface to maximum depth using a Li-Cor quantum sensor; the light extinction coefficient (K) was calculated from these data, as in Madariaga (1989).

Duplicate samples of 500–1500 ml for nutrient analyses and for total photosynthetic pigment determinations were subsequently filtered through glass-fibre filters (Whatman GF/C). Filters for pigment analysis were deep-frozen at –20° C and later homogenized and extracted for 24 h in 90% acetone. Absorbances were measured with a Shidmazu UV-24 spectrophotometer, and chlorophyll *a* concentrations were calculated using the trichromatic equations of Jeffrey & Humphrey (1975). No estimations were made for phaeopigments. Prefiltered samples for nutrient analyses were also kept frozen at –20° C in the dark, for a maximum period of 15 days, and later analyzed for inorganic nutrients (ammonia, nitrate, nitrite, phosphate, and silicate) according to the methods described in Strickland & Parsons (1972).

Rates of primary production were measured by the standard ^{14}C method (Steeman-Nielsen, 1952), and the distribution of ^{14}C in low molecular weight metabolites (LMWM), lipids, polysaccharides and proteins was determined using the extraction procedure of Li et al. (1980), as described by Madariaga & Fernández (1990). This method yields an aqueous methanol-soluble fraction (LMWM), a chloroform-soluble fraction (lipid), a hot 5% trichloroacetic acid-soluble fraction (polysaccharide), and a hot trichloroacetic acid-insoluble fraction (protein). Immediately after collection, samples were prefiltered through a 150 μm Nytal screen to eliminate larger zooplankton. Each sample was incubated for 3 h in 65-ml Nalgene polycarbonate bottles (two light and one dark for primary production measurements, and two light replicates for the distribution patterns of newly incorporated carbon), with 4 μCi $NaH^{14}CO_3$ in on deck-type incubators equipped with a refrigerated water flow circulator (FC 15 Grant Instruments Ltd.) and a thermostatic heater (FH 16D Grant Instruments Ltd.) to mantain temperature at *in situ* levels. In order to simulate light natural conditions, samples were exposed to different light regimes using neutral density screens. For each sampling site, a fraction of the ambient irradiance (100, 75, 50, 25, or 10%) was chosen according to the corresponding light extinction coefficient. This incubation system was tested by measuring the primary production of some duplicate samples from the central station which were incubated *in situ*. No significant differences were detected between the *in situ* samples and those in on deck type incubators (Madariaga & Orive, 1989). During the incubations, incident P.A.R. was recorded with a Li-Cor integrating quantum meter.

In addition, photosynthesis–irradiance (*P–I*) parameters were determined on samples taken at stations 2 and 4 following the procedures described by Madariaga & Joint (1992). Subsamples were incubated for 3–4 h at 24 different irradiances in an incubator cooled with circulating water at*in situ* temperatures. The incubation period was kept as short as practicable in order to reduce the photoadaptation which would occur in the light gradient.

All incubations for primary production determination were terminated by gentle filtration through 0.45-μm Millipore membrane filters, which were dried and counted in a Packard Tri-Carb 2000 CA scintillation counter, using the OptiScint 'HiSafe' (LKB Wallac) scintillation cocktail. Total CO_2 concentration was estimated from alkalinity measurements as described in Strickland & Parsons (1972). Total radioactivity for primary production estimates was compared with the sum of the activity of each macromolecular fraction and, based on 186 analyses from this study, the mean (± standard error, SE) recovery of activity after extraction was 98.5(± 1.9)%.

Diurnal variability of the photosynthetic characteristics and carbon metabolism of phytoplankton was studied on 15 July. Phytoplankton from waters corresponding to stations 2 and 4 (salinities about 27 and 11 PSU, respectively) were enclosed at dawn in duplicate 12-l acid-washed Nalgene polycarbonate bottles, and incubated *in situ* at station 2. Each bottle was filled from different Van Dorn bottle casts in order to achieve complete independence between duplicates. In all cases, the water was filtered through a 150-μm nylon mesh to remove larger zooplankton. Subsamples were taken from each bottle every 2 h in order to determine the chlorophyll *a* and nutrient concentrations, primary production, the distribution pattern of photosynthetically incorporated carbon, and phytoplankton composition. *P–I* parameters were also measured three times throughout the day.

Phytoplankton samples were preserved in Lugol's iodine solution, and later identified at ×100 and ×400 magnifications under an inverted Nikon microscope (Utermöhl, 1958).

Meteorological data were provided by the Meteorological Service of the Basque Country and the Oka river inflow variations by the Ibaizaintza service of the County Council of Biscay.

P-I curves were fitted to the hyperbolic tangent model suggested by Jassby & Platt (1976) using a SAS procedure (SAS Institute, 1985) on an IBM 9370 computer. Statistical analyses were performed using the Statview 5.0 program for a Macintosh personal computer.

Results

Meteorological conditions and hydrographic variability

Environmental conditions were highly variable throughout the study period (Table 1). Short frontal disturbances passed at intervals of 3–4 days through the study area which brought about very unstable climatic conditions. Several rain pulses were registered at the beginning of the survey, which were accompanied by relatively strong winds and low irradiance levels, and immediately altered river inflow rates. However, weather conditions became more sunny and calm towards the end of the survey. A transition from spring to neap tides also occurred during the study period.

Spatio-temporal variability in the estuarine hydrographic properties during the survey is summarized in Table 2. Strong salinity gradients were found when river discharge increased after rain pulses, but day-to-day changes in salinity occurred in response to both tidal and river runoff variations. Temperature was also related to climate conditions, and changed in response to the total irradiance. There was considerable vertical homogeneity in the water column. Increases in the light extinction coefficient were inversely correlated with salinity (correlation index $r = -0.902$, $n = 65$). Light attenuation, however, was not high enough to avoid PAR penetration down to the bottom. Dissolved inorganic nutrient concentrations were also inversely correlated with salinity ($r = -0.778$ to -0.976, $n = 65$) as freshwater pulses were the principal sources of nutrient inputs to the estuary. In consequence, concentrations of inorganic nutrients were high in the

Table 1. Temporal changes of meteorological conditions, river flow, and tidal amplitude during the survey

Day (July 1993)	Irradiance (E/m^{2}d)	Maximum wind (Km/h)	Rainfall (mm)	River flow (m^3/s)	Tidal amplitude (m)
1	22.74	16.08	8	0.31	2.84
2	6.35	12.88	42	0.83	3.14
3	22.75	5.08	0	0.56	3.39
4	20.47	6.54	0	0.40	3.52
5	15.48	18.54	10	0.35	3.49
6	41.37	8.13	12	0.41	3.30
7	55.02	8.17	0	0.38	3.00
8	52.73	8.58	0	0.31	2.76
9	40.92	11.29	0	0.29	2.34
10	25.02	12.42	25	0.49	1.94
11	47.28	14.04	4	0.44	1.58
12	51.82	7.21	1	0.38	1.29
13	55.46	6.50	0	0.32	1.16
14	54.09	9.00	0	0.28	1.24
15	50.46	6.96	0	0.24	1.54

Table 2. Mean values (± SE) of hydrodynamic parameters at each sampling station during the study period: salinity (PSU), temperature (° C), and the extinction coefficient, K (m^{-1}), n = 13; nutrients (μM), n = 26

Station	Salinity	Temperature	K	Ammonia	Nitrate	Nitrite	Phosphate	Silicate
ST 1	34.5 ± 0.2	19.0 ± 0.2	0.37 ± 0.03	2.0 ± 0.2	5.3 ± 0.8	0.13 ± 0.01	0.31 ± 0.02	2.5 ± 0.3
ST 2	26.8 ± 0.8	19.4 ± 0.4	1.20 ± 0.08	6.3 ± 1.0	14.2 ± 1.6	0.54 ± 0.04	0.46 ± 0.06	17.8 ± 1.5
ST 3	21.0 ± 0.9	19.7 ± 0.3	1.79 ± 0.07	21.8 ± 2.2	28.1 ± 2.4	0.98 ± 0.04	0.89 ± 0.09	37.4 ± 2.3
ST 4	10.8 ± 1.3	19.4 ± 0.5	2.13 ± 0.12	42.7 ± 4.5	48.2 ± 3.6	1.63 ± 0.05	3.01 ± 0.29	79.3 ± 3.9
ST 5	4.9 ± 0.8	18.9 ± 0.5	2.60 ± 0.20	68.3 ± 5.5	56.6 ± 4.1	1.68 ± 0.08	5.65 ± 0.52	105.5 ± 2.1

eutrophic upper reaches of the estuary, but relatively low in the neritic zone of the estuary.

Phytoplankton composition, biomass and production

Phytoplankton community composition of the Urdaibai estuary during this study was dominated by a diverse contingent of neritic diatoms with *Leptocylindrus danicus*, *Chaetoceros socialis* and *Cyclotella* as the most abundant taxa at the lower end of the estuary (Orive et al., 1998). In the middle and upper estuary, high proportions of detritus were found in some samples when river discharge increased. As a consequence it was not possible to identify small forms other than small flagellates, mainly cryptophyceans. These forms were very abundant in the estuary throughout the survey. As weather conditions stabilized towards the end of the study, coinciding with neap tides, high densities of a phytoflagellate-dominated community were found in the middle and upper reaches of the estuary. Dominant species were the dinoflagellate *Peridinium quinquecorne*, cryptophyceans and other small flagellates in the middle estuary, and *Euglena* sp., the dinoflagellate *Peridinium foliaceum*, and cryptophyceans in the upper estuary (Orive et al., 1998).

Phytoplankton biomass followed the same variation pattern (Fig. 2), with the highest chlorophyll *a* concentrations (up to 85 μg Chl *a* l^{-1}) measured under stable climatic conditions. Development of typical summer blooms in the upper estuary were observed after the initial freshets, but the occurrence of subsequent rain pulses temporarily limited phytoplankton growth. Semi-*in situ* carbon fixation rates at station 5 ranged from 3.2 μg C l^{-1} h^{-1} on day 3,

Table 3. Mean values (± SE) of physiological parameters of phytoplankton at each station ($n = 26$; p indicates the significance of the observed differences between stations using one-way ANOVA; SNK file shows the results of the Student–Neuman–Keuls test, $p < 0.05$; n.d.–not determined)

Station		$P^B{}_m$	α^B	I_k	P^B	480/665	% Met	% Lip	% Poly	%Prot
ST 1	Mean	n.d.	n.d.	n.d.	7.3	0.95	44.1	21.2	19.5	15.2
	SE	–	–	–	(0.7)	(0.02)	(1.9)	(1.0)	(1.6)	(1.1)
ST 2	Mean	10.2	0.034	298.3	7.9	0.88	39.5	24.1	15.5	20.9
	SE	(0.8)	(0.002)	(11.1)	(1.0)	(0.01)	(1.3)	(1.4)	(1.1)	(1.4)
ST 3	Mean	n.d.	n.d.	n.d.	7.2	0.81	34.3	24.1	15.9	25.7
	SE	–	–	–	(0.9)	(0.02)	(1.7)	(1.2)	(1.2)	(1.8)
ST 4	Mean	5.6	0.026	225.7	4.6	0.80	35.1	22.3	19.9	22.7
	SE	(0.4)	(0.002)	(10.6)	(0.5)	(0.02)	(1.9)	(1.3)	(1.2)	(1.5)
ST 5	Mean	n.d.	n.d.	n.d.	2.7	0.91	30.5	25.2	22.1	22.2
	SE	–	–	–	(0.3)	(0.04)	(1.8)	(1.3)	(1.7)	(1.3)
	p<	0.0001	0.0098	0.0001	0.0001	0.0001	0.0001	0.0001	0.0001	0.0001
	SNK	2>4	2>4	2>4	1=2=3>	1=2=5>	1=2>3=	1=2=3=	1=2=3=	1<2=3=
					4>5	3=4	4=5	4-5	4<5	4=5

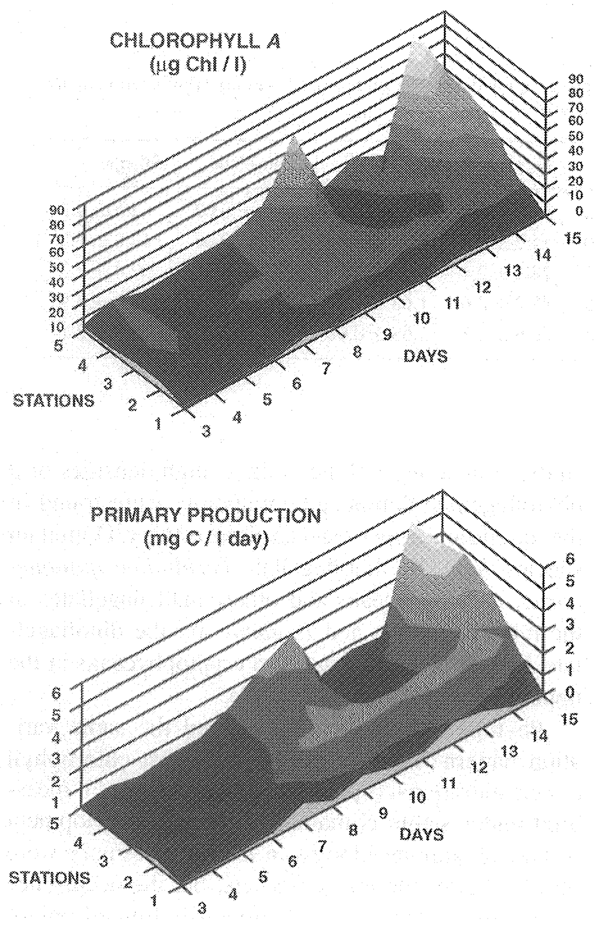

Figure 2. Spatio-temporal changes in chlorophyll *a* concentration and total primary production throughout the study.

to 353.7 µg C l^{-1} h^{-1} on day 14. As incubations for these determinations were performed at different times of day, total daily carbon fixation was calculated using Jassby & Platt's model for total daily irradiance values (Fig. 2), thus correcting the variability induced by the photoperiod (Madariaga, 1995). The spatio-temporal distribution pattern of carbon assimilation rates paralleled the changes observed in chlorophyll *a* throughout the survey, showing maximum rates during the phytoplankton bloom occurred at the end of the study period.

Physiological properties of phytoplankton

Changes of phytoplankton physiological parameters at each sampling station are summarized in Table 3. One-way analysis of variance (ANOVA) was used in order to test for differences in these parameters between stations. Multiple comparison between treatments (i.e., stations) was subsequently performed using the Student–Neuman–Keuls (SNK) test. Photosynthetic parameters derived from P–I curves at stations 2 and 4 showed significant differences. Assimilation numbers $P_m{}^B$, photosynthetic efficiencies α^B, and the derivative I_k ($P_m{}^B/\alpha^B$) values were always higher at station 2. The same trend was observed for photosynthetic rates per unit of chlorophyll P^B, with lowest values measured at the upper part of the estuary. During the study period, P^B measurements represent on an average 77.3 (±3.0)% of the assimilation numbers $P_m{}^B$, i.e., maximum productivity values under full irradiance (n=52). The ratio of light absorption at 480 and 665 nm by 90% acetone extracts of phytoplankton, a potential indicator of their nutritional status, was rather homogeneous along the estuary, but significantly lower values were found at stations 3 and 4.

Radioactive carbon labelling patterns also showed significant differences between stations. The percentage of carbon incorporated into LMWM fraction during the study showed a marked gradient along the estuary, with highest values in the neritic zone. However, the relative incorporation of ^{14}C into storage products, i.e., lipids and polysaccharides, did not show much variation, highest proportions corresponding to phytoplankton from the upper part of the estuary. The percentage of photosynthetic carbon incorporated into protein was significantly lower in the outer estuary. Figure 3 shows spatio-temporal changes in the patterns of newly incorporated carbon into macromolecules. In general, temporal variations seem to be more important than spatial variations. The relative incorporation of carbon into LMWM showed a high variability, and ranged from 14.5 to 62.4% (mean= 36.7; SE= 0.8; n= 130). Highest values were found at the beginning of the survey, but they diminished gradually as weather conditions improved. Estimates of the percentages of carbon fixed into lipids and polysaccharides varied between 12.3 and 39.2% (mean= 23.4; SE= 0.6; n= 130) and between 8.4 and 37.9% (mean= 18.6; SE= 0.6; n= 130), respectively. Both fractions showed highest values towards the end of the study period. Finally, the relative incorporation of carbon into proteins fell within the range of 7.1–52.0% (mean= 21.3; SE= 0.7; n= 130). Maximum percentages of protein labelling were found on day 7 at station 3, just after the rain pulse occurred on days 5–6.

Correlations of physiological parameters with environmental factors are showed in Table 4. Strong positive correlations were found between photosynthetic parameters and carbon fixation rates per unit of chlorophyll with the light received during incubation periods. In some cases negative correlations can also be observed with nutrients, mainly at station 1. The percentage of carbon incorporated into LMWM correlated negatively with light in all cases, and positively with nitrogenated compounds at several stations. On the contrary, positive correlations between the relative incorporation of carbon into lipid and light were statistically significant at stations 2, 3 and 4. The percentage of carbon fixed into polysaccharides also showed a positive correlation with light, but only at station 1. Significant positive correlations of this fraction were found with temperature at stations 2, 3 and 4, and also negative correlations with nutrients,

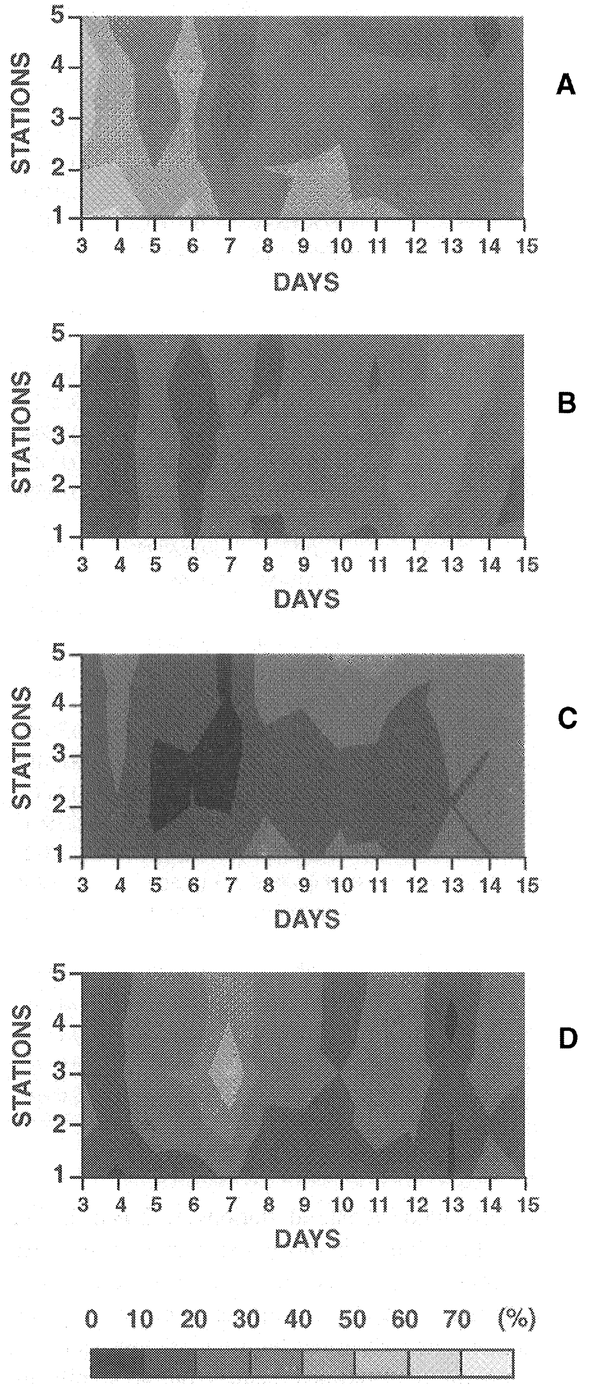

Figure 3. Spatio temporal changes in the percentage of photosynthetic carbon incorporated into macromolecules (A–LMWM; B–lipid; C–polysaccharide; D–protein).

Table 4. Correlation coefficients between physiological parameters and environmental factors during the survey at each station (* $p < 0.05$; ** $p < 0.01$; $n = 26$)

Station	Parameter	Light	Temperature	Ammonia	Nitrate	Phosphate	Sillicate
1	P^B	0.4615*	0.1857	–0.5845**	–0.5079**	0.5042**	–0.5891**
	% LMWM	–0.8014**	–0.2972	0.5112**	0.3112	–0.0026	0.7101**
	% LIPID	0.2528	0.0260	0.0069	–0.0784	–0.2077	–0.0374
	% POLYSAC	0.5160**	0.1425	–0.4323*	–0.2990	0.0975	–0.6951**
	% PROTEIN	0.4267*	0.2899	–0.2803	–0.0439	0.0568	–0.2119
2	P^B	0.6356**	0.2265	–0.1383	–0.3126	–0.1058	–0.0019
	$P^B{}_m$	0.6937**	0.0089	0.0043	–0.3844	–0.1166	–0.0919
	α^B	0.8031**	0.0389	–0.0690	–0.4557*	–0.1279	–0.0403
	% LMWM	–0.6588**	0.2254	–0.2295	0.2943	–0.2748	–0.1469
	% LIPID	0.5749**	–0.3004	0.1389	–0.1188	0.0775	0.2214
	% POLYSAC	0.2500	0.8585**	–0.6259**	–0.6264**	–0.6257**	–0.6851**
	% PROTEIN	–0.1211	–0.6158**	0.5921**	0.3319	0.6971**	0.4770*
3	P^B	0.6672**	0.2679	–0.2217	–0.4717*	–0.0233	0.0389
	% LMWM	–0.6574**	–0.0738	0.1484	0.3847	–0.0691	–0.1479
	% LIPID	0.6287**	0.0989	–0.1360	–0.3393	0.0981	0.1099
	% POLYSAC	0.2409	0.7176**	–0.3294	–0.6068**	–0.3012	–0.4158*
	% PROTEIN	0.0620	–0.4733*	0.1659	0.2562	0.2036	0.3489
4	P^B	0.5781**	0.5015**	–0.0168	–0.7704**	0.0259	–0.1932
	$P^B{}_m$	0.5579**	0.6219**	–0.0319	–0.7549**	–0.0393	–0.3778
	αB	0.4857*	0.3148	0.2887	–0.5592**	0.2220	–0.1466
	% LMWM	–0.6893**	–0.3864	0.2063	0.5288**	0.0242	0.2181
	% LIPID	0.5179**	0.2572	0.2141	–0.3741	0.3496	–0.0223
	% POLYSAC	0.2822	0.5086**	–0.1360	–0.8045**	–0.1182	–0.3494
	% PROTEIN	0.2071	–0.1269	–0.3314	0.2759	–0.2316	0.168
5	P^B	0.4749*	0.2387	–0.0083	–0.2207	0.1966	–0.2855
	% LMWM	–0.4899*	–0.1158	–0.0059	0.6695**	–0.3301	–0.1875
	% LIPID	0.2732	0.1135	0.2859	–0.3569	0.5211**	0.3113
	% POLYSAC	0.1781	0.0872	0.0511	–0.4173*	0.1662	0.0060
	% PROTEIN	0.1662	–0.0648	–0.3284	–0.0208	–0.2663	–0.0527

particularly with silicate at stations 1, 2 and 3. The relative allocation of photosynthetic carbon into protein correlated positively with light at station 1, and with nutrients at station 2. Significant negative correlations were also observed between protein labelling and temperature at stations 2 and 3, but no significant relationship was found at stations 4 and 5.

Diurnal photosynthesis and carbon metabolism patterns

Studies of diurnal variability in the photosynthetic capacity and carbon metabolism of phytoplankton were conducted on 15 July 1993. Biomass concentration of the phytoplankton enclosed in microcosms exhibited the expected dynamics of bloom formation and collapse after nutrient depletion (Fig. 4a). Nutrient concentrations displayed similar patterns for both water samples, and decreased as a function of phytoplankton growth. Ammonia levels fell before nitrate and phosphate depletion, which occurred at the end of the day. Silicate concentrations also diminished but no depletion was measured. As weather conditions were optimal, full irradiance levels were recorded during all day. Photosynthetic rates per unit of biomass (P^B) tracked light intensity very closely (Fig. 4b and Table 5).

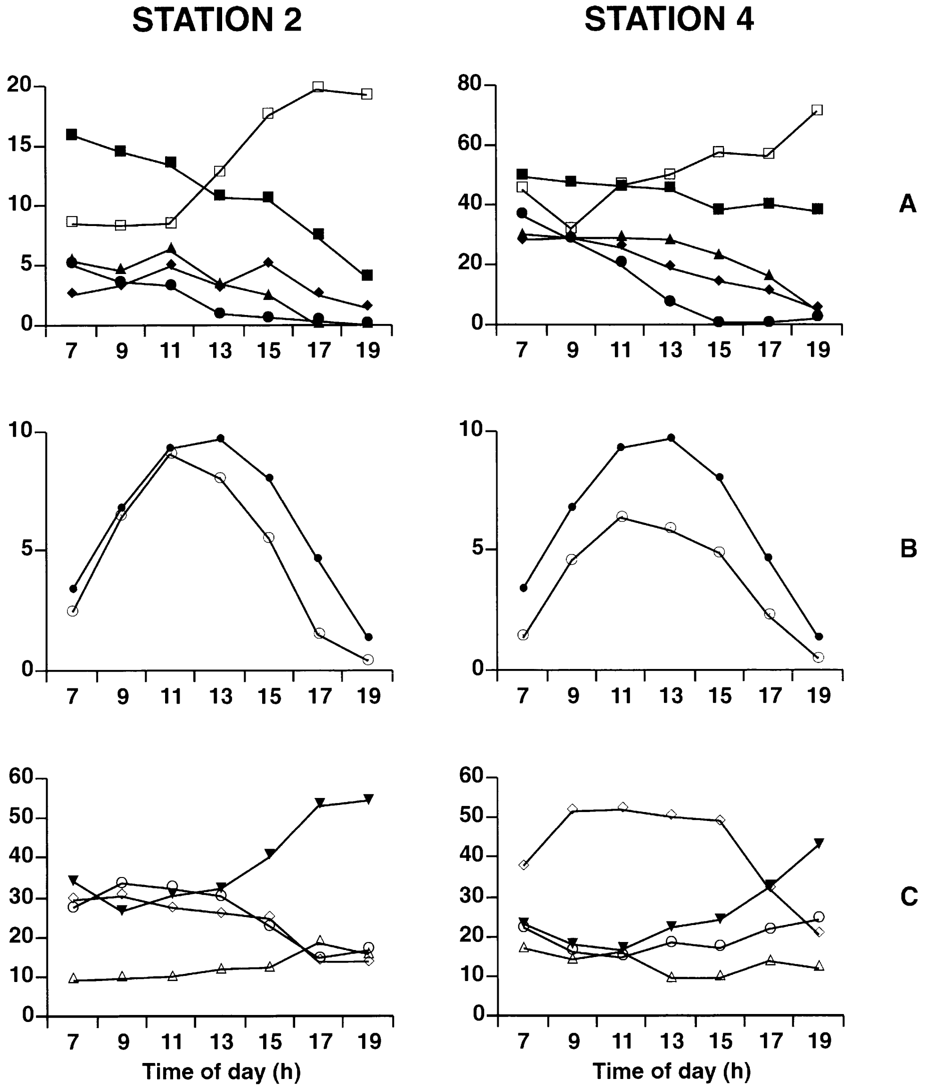

Figure 4. Results of the phytoplankton diurnal variation experiment: (a) Changes in chlorophyll *a* (□, μg Chl l^{-1}) and nutrient concentrations (μmol l^{-1}: ■, silicate; ●, ammonia; ▲, nitrate; ♦, phosphate, ×10) in the experimental bottles; (b) incident PAR (•, ×200 μE m^{-2} s^{-1}) and productivity (⊙, μg C (μg Chl *a*)$^{-1}$ h^{-1}) values throughout the day; (c) percentage of photosynthetic carbon incorporated into ▼, LMWM; ◇, lipid; ○, polysaccharide; and △, protein.

The percentage incorporation of inorganic carbon into macromolecules and LMWM was different at the two samples, but temporal changes were similar in both water masses (Fig. 4c). Under light and nutrient sufficient conditions carbon incorporation patterns kept rather constant, but towards the end of the experiment, when light levels decreased and nutrients were limiting for phytoplankton growth, the percentage of carbon assimilated as LMWM increased while lipid labelling decreased. The relative incorporation of carbon into polysaccharides also diminished in the station 2 sample, but slightly increased at the end of the experiment in the station 4 sample. Protein labelling was rather constant throughout the day.

Discussion

One of the most important questions concerning phytoplankton ecophysiology is to know how the cells adjust their metabolism to different environmental conditions and to assess the effects – both direct and indirectly – of these changes at various levels within the ecosystem. To this effect, the study of the factors

Table 5. Correlation coefficients between physiological parameters and environmental factors during the diurnal experiment (* $p < 0.05$; **$p < 0.01$; $n = 14$)

Station	Parameter	Light	Ammonia	Nitrate	Phosphate	Silicate
2	P^B	0.9546**	0.2676	0.7045**	0.7629**	0.5503*
	% LMWM	–0.6526**	–0.7180**	–0.9136**	–0.5255	–0.8864**
	% LIPID	0.5112	0.7854**	0.9195**	0.5321	0.9407**
	% POLYSAC	–0.3825	–0.8056**	–0.9319**	–0.4460	–0.8635**
	% PROTEIN	0.6563*	0.6902**	0.9103**	0.4790	0.8187**
4	P^B	0.9906**	0.0158	0.6538*	0.4452	0.2637
	% LMWM	–0.7527**	–0.5808*	–0.9617**	–0.8837	–0.7197
	% LIPID	0.8921**	0.3122	0.8590**	–0.8560**	0.4914
	% POLYSAC	–0.2473	0.7542	0.2912	–0.5928	0.6175
	% PROTEIN	–0.9022	–0.2053	–0.7018	–0.5928	–0.3442

that regulate photosynthesis and carbon metabolism is crucial. Patterns in the relative synthesis of cell compounds can be associated with different environmental and physiological conditions, but few attempts have been made so far to study such relationships in estuarine ecosystems, probably because the interactions usually occurring between environmental factors, and the interspecific differences that characterise natural assemblages renders this effort difficult.

Day-to-day variability in the hydrographical and biological features of the Urdaibai estuary during this study was due to changes in meteorological and tidal conditions. At the beginning of the survey, low atmospheric pressure systems moved over the study area at irregular intervals, and brought short pulses of rain that significantly altered river flow rates. Variations in river runoff determined not only the hydrological zonation of the estuary, but also water turbidity, nutrient loading and temperature conditions. Towards the end of the study period, however, the atmospheric situation stabilised, and therefore, environmental conditions for phytoplankton growth improved. Favourable light and temperature levels, and longer flushing times due to reduced river inflow and neap tidal conditions resulted in the development of a mixed phytoflagellate-dominated community in the upper part of the estuary. Summer phytoplankton blooms like this are rather common in shallow turbid estuaries of temperate latitudes, and can be explained by variations in both the tidally regulated mixing regime (Fichez et al., 1992; Lizon et al., 1998), and the upper watershed rainfall, which subsequently regulates downstream river flow (Mallin et al., 1993; Zakardjian et al., 2000).

Previous studies carried out in this ecosystem (Madariaga & Orive, 1989; Madariaga et al., 1989, 1992) showed that phytoplankton biomass and production in the upper estuary was low during seasonal periods of high river flow and short residence time, but highly increased during summer seasons of low river flow and long residence time. During this study, under unstable environmental conditions, phytoplankton biomass and primary production varied gradually over a 3–4-day period, but shorter scales of variation (diurnal and day-to-day changes) became more important under stable, growth favourable conditions. This shift in the dominant period (time scale) of the variability associated to phytoplankton dynamics agrees with the general pattern described by Sinclair et al. (1981). In general, temporal changes in phytoplankton were more important than spatial variations along the estuarine longitudinal axis.

Changes in the photosynthetic characteristics of phytoplankton during this study were discussed in a previous paper (Madariaga, 1995). P^B, $P_m{}^B$ and α^B values were lower in the upper, more turbid zone of the estuary, this denoting photoadaptation of phytoplankton to a lower light regime (Harding et al., 1986). In the neritic zone, however, photosynthetic parameters were always higher because water turbidity is always very low (Madariaga, 1989). On the other hand, short-term (day-to-day) temporal variations in photosynthetic parameters appeared to be consistently correlated with irradiance as they are specific to the time-scale of change in light conditions (Falkowski, 1984; MacIntyre et al., 2000).

Nevertheless, in some stations significant relationships were found with nutrient availability and temperature, which could also be important in controlling the photosynthetic capacity of phytoplankton populations (Campbell & Bate, 1987; O'Donohue & Dennison, 1997). While temperature seems to have an effect on photosynthetic parameters on a longer time-scale (Côte & Platt, 1983), in the case of nutrients, the concentration in the environment is not as important as the intracellular concentration (Heath et al., 1990). According to these authors, the ratio of light absorption at 480 and 665 nm by acetone extracts of phytoplankton pigments may be a useful indicator of nutritional status of natural phytoplankton populations. The absolute values of this ratio cannot be used to predict nutrient limitation, but can be used as a relative index to detect nutrient limitation (Schlüter et al., 1997). In this study, the 480/665 ratio was always very low, and did not show significant changes, thus confirming that nutrients are not usually limiting in the Urdaibai estuary (Madariaga & Orive, 1989; Madariaga et al., 1994).

Partitioning of photosynthetically incorporated carbon among macromolecular fractions in this study also reflected ambient light and nutrient conditions in the estuary, and changes in the metabolic patterns were associated with spatial and temporal gradients. The highest proportion of carbon was incorporated into the LMWM, specially in the lower estuary. This spatial pattern can be explained by both environmental factors and species-specific differences (Madariaga, 1992; Smith et al., 1997). In fact, some authors have shown that dinoflagellates have less incorporation of photosynthetically fixed carbon into LMWM than in diatoms and chrysophycean flagellates (Hitchcock et al., 1986). On the other hand, LMWM labelling was highly coupled to low irradiance conditions in all the sampling stations. High carbon fixation into LMWM is frequently associated with growth limitation by various environmental factors due to reduction in the rates of macromolecules synthesis and, therefore, slow utilisation of intermediate compounds in the metabolic pool, rather than to a fast rate of production. This fact would indicate a slow-growing phase of phytoplankton (Madariaga, 1992) under unfavourable environmental conditions.

The relative incorporation of carbon into storage materials, i.e., lipids and polysaccharides, also indicated differences in the species composition and the physiological role these compounds have under a fluctuating environmental regime. In general, there is an inverse relationship between carbon incorporation into storage products and LMWM. Thus, the rise of light and temperature that occurred at the end of the study period was accompanied by an increase in the percentage of carbon incorporated into lipids and polysaccharides, as shown by the positive correlations between these parameters. However, no consistent trends were found in all stations. The relative allocation of phytoplankton photosynthate to lipids tended to be related with incubation irradiance, while carbon assimilation into polysaccharide was more related to temperature and nutrient availability. Discrepancies may be due to different algae, which may exhibit different physiological responses. Moreover, in complex natural communities, there is always the possibility that environmental variables such us temperature or light intensity are acting as proxies of other controlling variables. Therefore, empirical correlations between photosynthate allocation and abiotic environmental factors may be of limited generality (Wainman & Lean, 1996, 1997).

No clear pattern of reallocation of photosynthetic carbon into the protein fraction appeared in this study. Highest percentages were measured at the beginning of the bloom that developed in the middle and upper estuary, but overall, they were rather constant throughout the survey. It is well known that environmental stress, such us limited light or nutrient availability, causes cells to conserve protein synthesis. Konopka & Schnur (1980) showed that in strong light, the rate of carbon fixation exceeds the rate of nutrient assimilation, thus the excess of carbon derived from photosynthesis is stored as polysaccharides. Inversely, in low light the rate of carbon fixation and ATP production is light limited, so that the amount of carbon and energy produced does not exceed what can be assimilated into proteins. Similarly, Marañón & González (1997) argued that the maintenance of protein synthesis under low irradiance conditions and the enhancement of lipid synthesis when an excess of energy is available represent a general strategy of energy utilization by marine phytoplankton. It seems, however, that the pattern of carbon allocation into polysaccharide or into lipid under these conditions is species-specific in natural populations (Madariaga, 1992; Smith et al., 1997). The statistical relationships obtained in this study corroborate these previous results.

It must be pointed out, however, that results from this study were obtained from samples incubated during light periods. Such incubations avoid the tracer appearing increasingly in organisms and pools other than algae (Smith et al., 1984), but cannot demonstrate any further reallocation that might occur during a day–night cycle. In fact, laboratory cultures and field studies have shown that carbon incorporation into protein continues during the dark period, which is sustained by carbon and energy from other fractions (Hama et al., 1988; Madariaga & Joint, 1994).

Diurnal patterns of photosynthesis and carbon metabolism are also very important to analyse short-term changes of phytoplankton physiological state as this diurnal variability is superimposed on the day-to-day variability (Maurin et al., 1999). P^B rates determined in the diurnal variability experiment tracked light intensity very closely, as observed previously (Marra & Heinemann, 1982; Jones et al., 1990). However, an absence of a marked diurnal variability in the phytoplankton photosynthetic capacity was observed, which allows calculation of diurnal primary productivity rather accurately from a single midday estimate of P–I parameters (Madariaga, 1995). On the other hand, the partitioning of carbon among biochemical fractions showed great diurnal variability. It is not clear whether these changes, that occurred during the second half of the day, are related to the diurnal periodicity of light-field (Jones et al., 1990) or to the effect of nutrient depletion within microcosms (Madariaga & Fernandez, 1990; Madariaga, 1992), as a consequence of the high initial concentrations of phytoplankton biomass. In any case, results from this experiment confirm those patterns in the photosynthetic carbon metabolism of phytoplankton observed during the survey.

The final aim of this study was to test previous models for environmental control of the physiological state of phytoplankton in the Urdaibai estuary. 'Physiological state' is a rather vague term which implies knowledge – both qualitative and/or quantitative – about growth rates and their possible reduction relative to a potential maximum due to a limiting factor. The maximum growth rate of phytoplankton is variable and is solely determined by physical parameters such as temperature, whereas the reduction in the growth rate can be caused by a limiting factor such as nutrient or light availability. Experiments carried out by Droop (1974) showed that for a single species only one factor can be limited at a time. In natural populations, however, photosynthesis and allocation of photosynthate, and thus the food quality and nutrient stoichiometry of the primary producers, appeared to be under complex control that could not be precisely predicted from any single environmental variable. The results obtained in this study show that the partitioning of photosynthetic end products is simultaneously affected by biotic and abiotic factors, although short-term light fluctuations may have a major influence on the physiological state of phytoplankton in the Urdaibay estuary.

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E. Orive, M. Elliott & V.N. de Jonge (eds), Nutrients and Eutrophication in Estuaries and Coastal Waters.

Evidence of a planktonic food web response to changes in nutrient input dynamics in the Mar Menor coastal lagoon, Spain

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Key words: coastal lagoon, eutrophication, pelagic food web, top-down control, jellyfish, size biomass-spectra

Abstract

Nutrient input dynamics in the Mar Menor coastal lagoon has recently changed as a consequence of changes in agricultural practises. An interannual comparison of the environmental variables and the planktonic biomass size-spectra was performed between 1988 and 1997. While nitrate concentration was low in 1988, the values in 1997 increased considerably. Since 1995, two alloctonous jellyfish species (*Rhyzostoma pulmo* and *Cotylorhiza tuberculata*) occurred in large numbers in summer time and reached peak abundance in summer of 1997. The size-spectra analysis comparison revealed that, in spite of changes in nutrient input that stimulated the growth of larger phytoplankton cells, there were no significant differences in the spectra slope which followed a similar seasonal trend in both years. However, the plankton biovolume considered under the size range compared (between 2 and 1000 μm diameter) was, paradoxically, always lower in 1997. Given that there were higher nutrient levels in 1997, this finding suggest a strong top-down control mechanism of size structure. Gut contents of jellyfishes showed their preference for large diatoms, tintinnids, veliger larvae and copepods, corroborating that size structure in these assemblages can be subject to top-down control. The implication of these results is that the feeding activities of large gelatinous zooplankton (jellyfishes) may play an important role controlling the consequences of eutrophication within the Mar Menor coastal lagoon.

Introduction

Nutrients are considered the main limiting factor for phytoplankton growth in shallow temperate ecosystems, where photon flux through the water column does not impose restrictions to photosynthetic requirements across seasons. The response of planktonic food webs to nutrient enrichment in coastal marine ecosystems varies widely world wide due to the broad range of both direct and indirect effects of the eutrophication process (Kerfoot & Sih, 1987; De Angelis, 1992; Scheffer, 1998). When a bottom-up control exists, the general patterns described include the substitution of macrophytes by macroalgae at the benthic level in a first step and then a change to a phytoplankton based system with anoxic events originated when light penetration is severely limited by phytoplankton absorption (Niehnius, 1992). However, the response of ecosystems to nutrient load increase differs widely because biological control mechanisms of the eutrophication process are not always the same. Predation can be a very efficient control mechanism providing alternative energy flow pathways in the food web by removing excess biomass generated by excess nutrients.

Organisms fit themselves to the environmental conditions determining the planktonic structure. Changes in taxonomic composition related to the eutrophication process influence the size structure (Sprules & Knoechel, 1984; Sprules & Munawar, 1986). Theory and natural patterns across ecosystems (Awagin et al., 2000) state that small phytoplankton cells dominate in temperate oligotrophic waters whilst large ones are comparatively more abundant in colder and rich nutrient waters. The implications of cell size for nutrient uptake (see review by Chisholm, 1992), pointed out the competitive advantage of small cells in poor nutrient water due to their higher surface to volume ratio which allows more efficient uptake and diffusion of nutrients. Although small cells also grow under high nutrient regimes, large cells outcompete them due to competitive advantages derived from larger size, such as better light adaptation (Geider et al., 1986) or predation avoidance (Raymont, 1963).

Abundance and size of cells determine both quantity and kind of potential predators, as grazing rates for most planktonic heterotrophs are size dependent (e.g. Frost, 1972; Bartram, 1980; Peters, 1994). In this way, changes in the size structure of phytoplankton assemblages may, therefore, induce changes in zooplankton population regulating the increase of different size organisms and, at the same time, size-dependent complex interactions in the food web determine, in turn, most of the changes in the plankton structure. However, the mechanisms and processes (including size-dependent relationships) involved in algal blooms or in the massive proliferations of jellyfishes are something not yet well understood.

The objectives of this paper are (a) to examine changes in the structure and size spectrum of the planktonic community in relation to differences in the nutrient loads, and (b) to asses the effect of the proliferation of jellies on the pelagic food web structure. To do this, we compare the plankton size structure in the Mar Menor coastal lagoon before and after the nutrient input regime changed over a 9-year period between 1988 and 1997, which lead into a massive proliferation of jellyfishes.

Description of the study area

The Mar Menor is a hypersaline coastal lagoon with a surface area of about 135 km^2. It is located in a semiarid region (113–498 mm mean annual rainfall) at the Southeast of Spain (37° 44′ N, 0° 47′ W) on the Mediterranean coast (Fig. 1). The mean depth is 3.5 m with a maximum depth of 6 m. The salinity of the lagoon waters ranges between 42 and 47 psu, due to high evaporation and low exchange rates with the Mediterraean. Water temperature ranges from 10 °C in winter to 31 °C in summer. No stratification exists in the water column. The Mar Menor is one of the largest coastal lagoons on the Mediterranean coast supporting a wide range of uses, including large tourist resorts and intensive agriculture. It is isolated from the Mediterranean Sea by a 22 km long sandy bar (La Manga), crossed by five very shallow channels. In the early 1970s, one of these channels (El Estacio) was dredged and widened to make it navigable. Since then, it has become the main connection with the sea and the average water residence time in the lagoon is about 0.79 years (A. Pérez-Ruzafa, 1989).

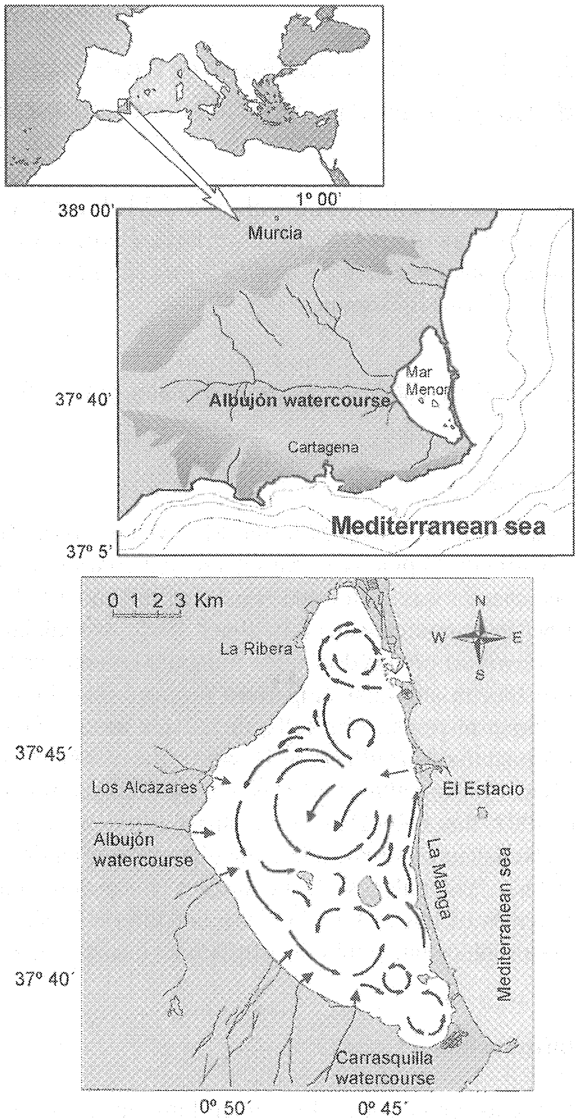

Figure 1. Location of the Mar Menor lagoon with main watercourses on the watershed and circulatory model.

In the 1970s, primary productivity was mainly benthic and the main macrophyte was the phanerogam *Cymodocea nodosa*. During the early 1980s, after the enlargement of El Estacio, the bottoms were covered by a mixed meadow of *Cymodocea nodosa-Caulerpa prolifera*, which biomass was about 280 g dw m^{-2} (I.M. Pérez-Ruzafa, 1989; Terrados & Ros, 1991). By the mid 1980s, a dense bed of the invasive macroalga, *Caulerpa prolifera*, covered most of the bottom, restricting the seaweed *Cymodocea nodosa* to small patches in shallowest areas. The high benthic macrophyte biomass contrasted with the low phytoplanktonic density (Ros & Miracle, 1984) and the oligotrophy of the waters (Gilabert, 1992).

Several watercourses drain into the lagoon (Fig. 1) but only become functional under episodic rainfall events as high evaporation and infiltration rates prevent waters to reach the lagoon under normal conditions (Lillo, 1978). However, during the last 10 years agriculture in the watershed has changed from extensive dry crop farming to intensively irrigated crops. Water for irrigation is diverted from rivers to the North (Tajo–Segura rivers diversion). These changes led to a rise in phreatic levels, as the overexploitation of ground water decreased. Some watercourses, such as El Albujón and La Carrasquilla, now maintain a regular flux feed by ground water with high nitrate levels to the lagoon. At the same time, increasing touristic activities lead to a rise in urban sewage which usually is the main input of phosphate in Mediterranean coastal lagoons (Vaulot & Frisoni, 1986) however, meanwhile in 1988 no urban wastewater treatment plants were installed, in 1997 they exist for the main cities, as La Ribera and Los Alcázares.

In the mid 1980s, two allochtonous species of jellyfish (*Rhyzostoma pulmo* and *Cotylorhiza tuberculata*) entered into the lagoon from the Mediterranean (A. Pérez-Ruzafa, 1989). After an initial period of slow growth, their population grew up to reach considerably greater quantities (Pérez-Ruzafa et al., 1997, technical report), resulting in serious inconveniences to tourism.

Materials and methods

Results obtained from two sampling programmes carried out in 1988 and 1997 are compared in this study. Figure 2 shows the location map of the stations sampled weekly in 1997. Environmental variables (temperature, salinity, nutrient concentrations, i.e. nitrate and phosphate), were measured at all stations. In addition, plankton samples were taken at sampling stations 2, 5, 10 and 18. In 1988, only station 2 was sampled.

Water samples for environmental variable measurements were taken at 1 m depth with a Niskin bottle. Samples for nutrient analysis were kept in dark and at 4 °C in the field and stored at −28 °C. Nitrate (NO_3-N) was determined following the method described by Parsons et al. (1984). Phosphate (PO_4-P) was analysed following the method described by Fernández et al. (1985) in 1988, whilst the method described in Parsons et al. (1984) was used during 1997. For salinity determination, a Beckman RS 7B salinometer was used in both years.

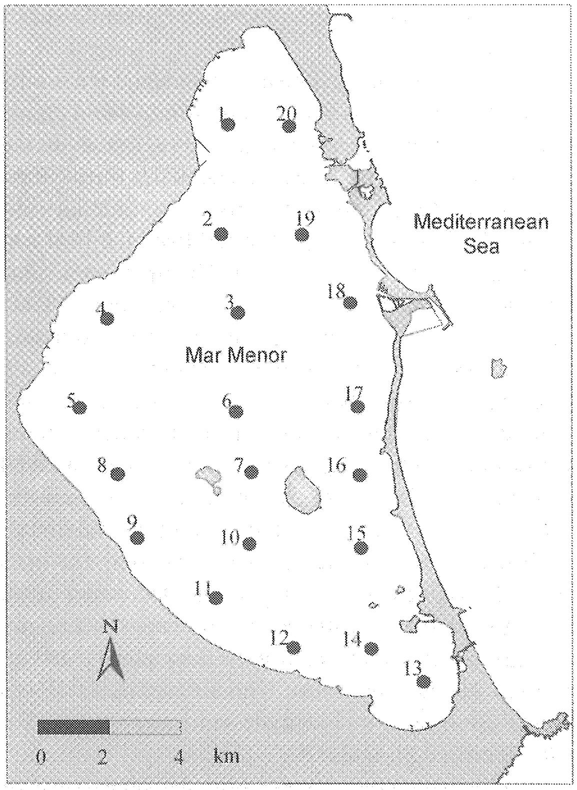

Figure 2. Map showing the location of the sampling stations used in this study.

Field sampling for plankton composition and size structure study was described in Gilabert et al. (1990). A sub-sample of 250 ml obtained with Niskin bottle was stored in a dark glass bottle and preserved with Lugol acetic solution to study nanoplankton (2–20 μm of Estimated Spherical Diameter) and microplankton (20–200 μm ESD). To estimate the abundance of the larger fraction of microplankton, 5 l water sample was filtered through a 45 μm gauze. The retained material was preserved in 5% formaldehyde. Finally, two horizontal tows using a Bongo (100 μm gauze) and a Juday-Bogorov (250 μm gauze) net, both equipped with flow meters, were carried out to estimate the abundance of the mesoplankton (>200 μm ESD). An extra net of 500 μm gauze was added in 1997 to count and measure fish larvae, ephyrae and juvenile stages of jellyfishes.

Census for adult jellyfishes abundance were only performed in 1997 as for 1988 no *Rhyzostoma pulmo* neither *Cotylorhiza tuberculata* were found at the sampling station. Adult jellyfish densities were estimated at the 20 sampling stations by combining simultaneous visual census performed from the boat and towed nets at a velocity of 1 not. The maximum value obtained for each sampling was considered as the more realistic data of the population. Several individuals were taken and measured in laboratory to obtain size frequency distributions. For gut content analysis, individuals were taken directly from the water column with a polyethylene bag; they were externally washed and carefully fixed with 5% formaldehyde-sea water solution in another bag. In the laboratory, individuals were dissected and gastric cavity and oral channels washed carefully with sprayer distilled water. Bag contents as well as washed materials were sequentially sieved through 1 mm (mainly for body pieces) and 56 μm (for gut contents) sieve. The filtrated material was preserved in 5% formaldehyde-sea water solution for later microscopic analysis.

In the laboratory, to estimate the abundance of nanoplankton and microplankton smaller than 45 μm, 100 ml of the first sub-sample were allowed to settle for 48 h. A variable area was scanned with a Nikon inverted microscope (×450); the scanning criterion was to count at least 300 cells of the most abundant form. To estimate the abundance of microplankton larger than 45 μm, material retained on the 45 μm gauze was resuspended and then allowed to settle for 24 h. The sedimented material was examined at ×150 until at least 100 individuals of the most abundant organism were counted. Finally, a sub-sample from the plankton nets was examined at ×40 with a similar scanning criterion to estimate mesozooplankton abundance.

Organisms were measured using non-destructive techniques with semiautomatic image analysers (Kontron IBAS-1 in 1988 and software Image Tool v 2.7 on a PC connected through a video-camera to the same inverted microscope, in 1997). Following biometrics parameters were measured: (i) major diameter (D), i.e. the longest distance between the two extremes of the organisms, and (ii) minor diameter (d), i.e. the width of the mean cross section (longest perpendicular distance between two points at the edge of the organisms). The organisms were considered as approximately ellipsoidal and the individual volume (V) was calculated as the 'equivalent ellipsoidal volume' ($V = (\pi \cdot \mathrm{d}^2 \cdot D)/6$). The calculated volumes for the overall size range were classified in a doubling ($\log_2$) scale of individual volume (so, upper limit of size classes correspond to 1, 2, 4, 8, 16 μm^3, etc). The common output for the overall size range was then a frequency distribution of counts. Biovolume per size class can be obtained by multiplying the density by the nominal size of the class. Following Platt & Denman (1977, 1978) a normalised – to the width of the size class - biomass function was used. Assuming that all the individuals within a size class are of similar size, and taking advantage of the octaval scale, the normalised biomass spectrum approaches a density spectrum. This procedure can be considered as standard for biomass spectra analysis (Rodríguez & Mullin, 1986; Sprules & Munawar, 1986; Gaedke, 1992; Blanco et al., 1994). Obviously, the size range of the organisms studied does not cover the whole planktonic community as very small (<2 μm) cells were not considered for the analysis due to the microscopic technique used and incomplete sedimentation in Utermöhl chambers and preservation problems.

Four size spectra corresponding to 11 May, 15 June, 14 July and 10 August for station 2 in 1988 were compared with other four spectra corresponding to 12 May, 16 June, 14 July and 11 August for station 10 in 1997.

Results

The comparison between nitrate and phosphate distribution along the year at station 2 (see Fig. 3) shows that while nitrate concentration in 1988 (Fig. 3a) was always under 1 μg-atm N-NO$_3^-$ l^{-1}, much higher concentrations occurred in 1997 (Fig. 3b), particularly during spring and summer (just at the harvest time when larger amounts of fertilisers are used in the lagoon's watershed). The results suggest that nitrate was mainly entering into lagoon via runoff in 1988 since a positive and significant correlation was found between nitrate concentration and accumulated rainfall during the week prior to the sampling day (r=0.45, n=21). Samples with concentration below the detection limit of the method were not included here. Furthermore, higher nitrate concentrations are usually found in the west coast of the lagoon, close to the mouth of the main watercourses, meanwhile lower concentrations are associated to the west coast and El Estacio channel influence area (Fig. 4). All these facts suggest that nitrate was related to the agriculture activity.

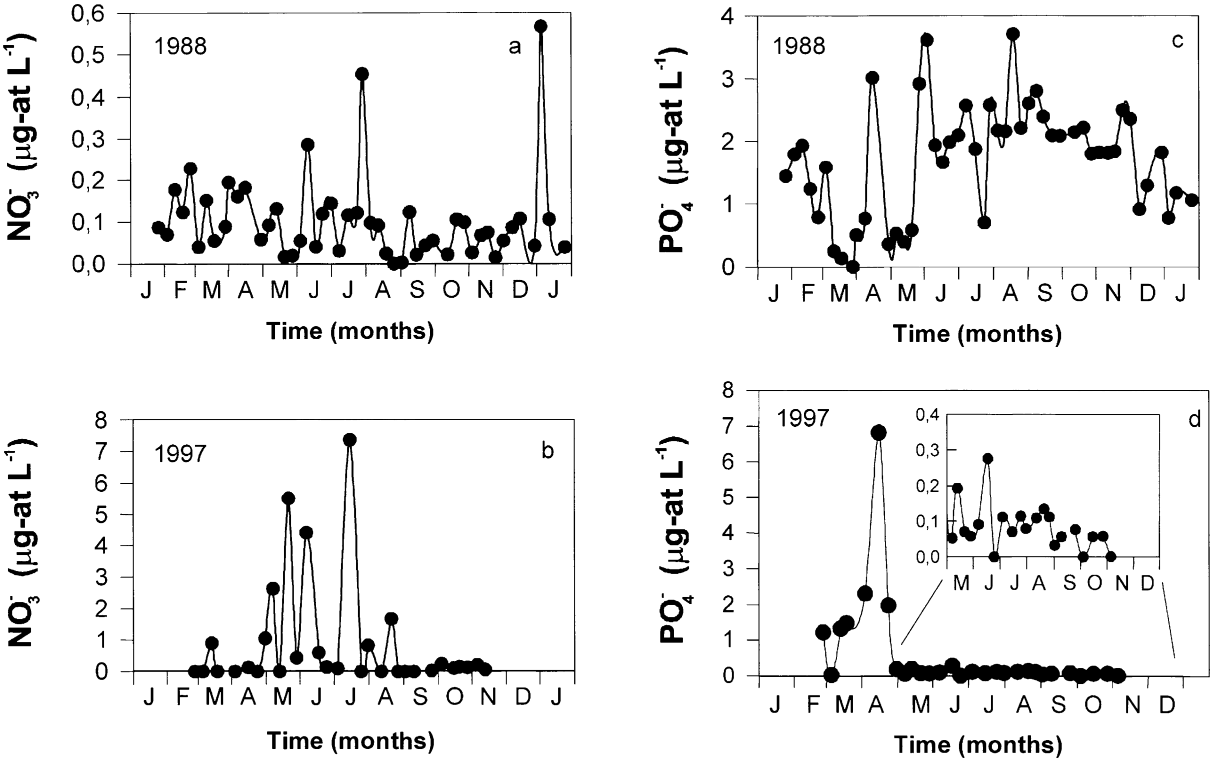

Figure 3. Temporal variation in nutrient concentration at station 2 in 1988 and 1997. (a) Nitrate in 1988, (b) nitrate in 1997, (c) phosphate in 1988 and (d) phosphate in 1997.

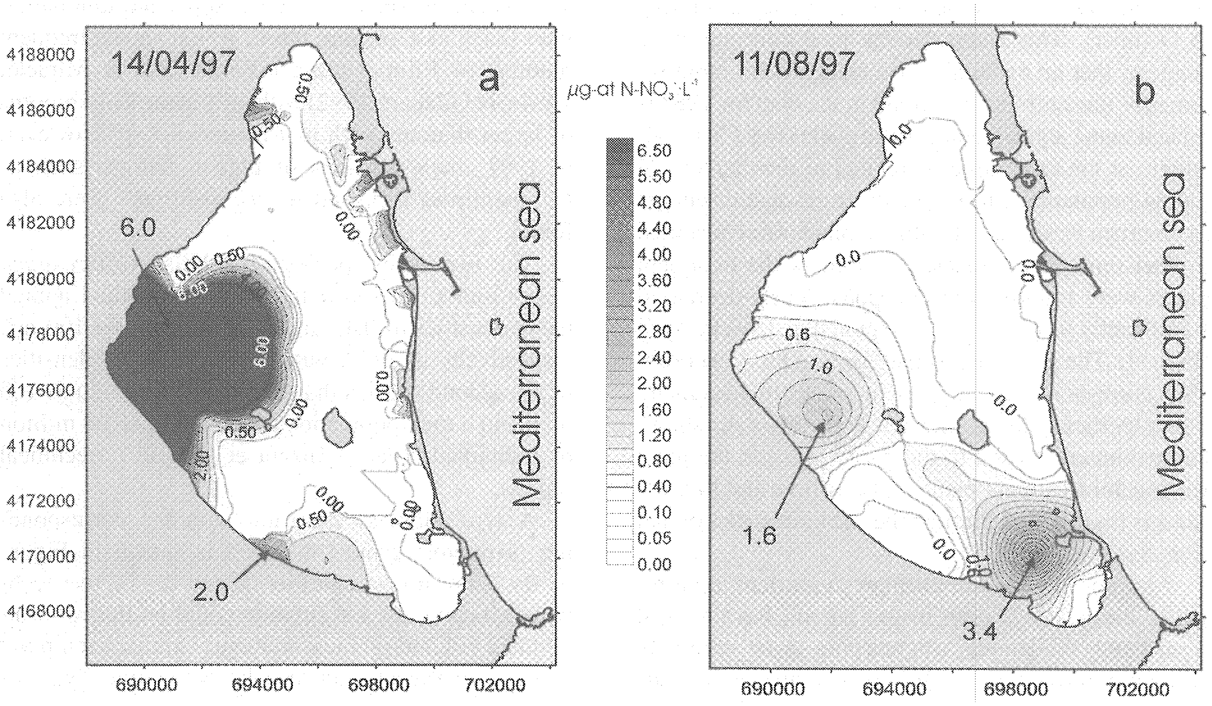

Figure 4. Spatial distribution of nitrate at the Mar Menor lagoon in April and August of 1997.

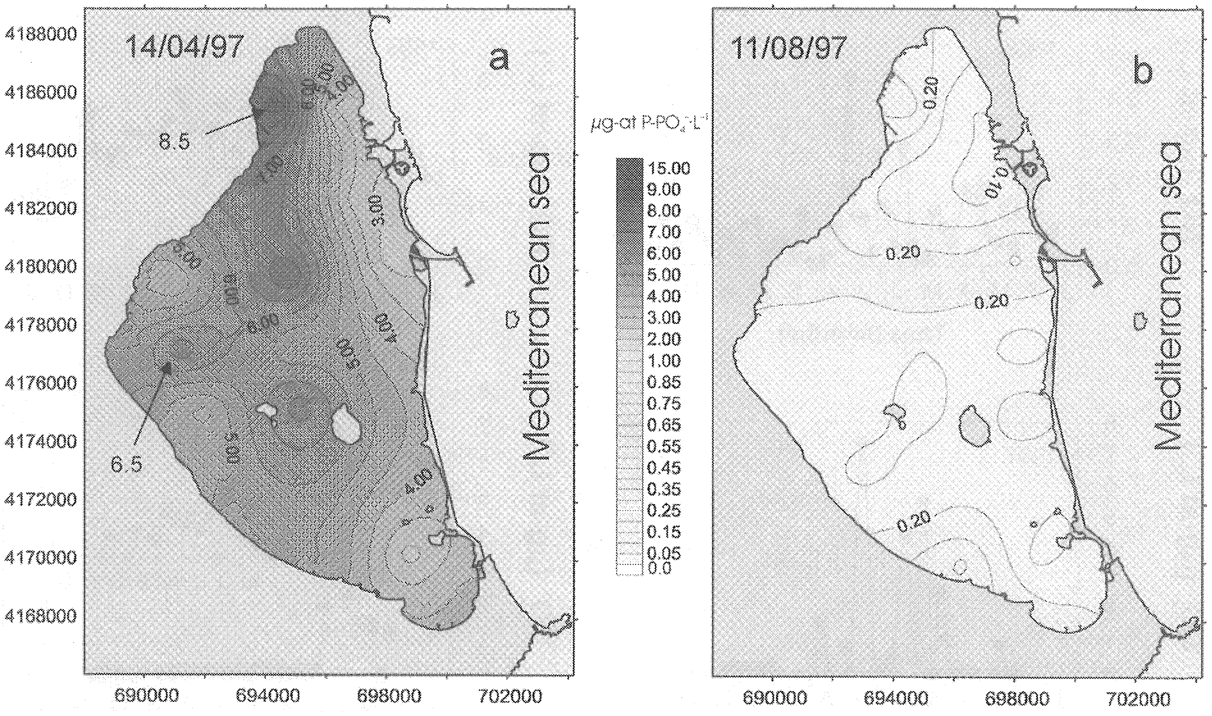

Figure 5. Spatial distribution of phosphate at the Mar Menor lagoon in April and August of 1997.

Drastic change in phosphate levels were found between 1988 (Fig. 3c) and 1997 (Fig. 3d, from May to October). This sharp decline in P concentrations suggests that an external cause changed the input regime for this nutrient. The trend in the data for 1988 is related to the annual temperature cycle (r=0.53, n=44) which, at the same time, is related to the increase of tourist population and indirectly to sewage discharge as no urban wastewater treatment plants were installed by that time. Nevertheless, as soon as the treatment plants were installed the phosphate levels decreased significantly in 1997, except in punctual dates in April (Fig. 3d). The peak observed in April of this year probably corresponds to failures in waste management. The superficial distribution or P in this date shows the maximums closed to the waste pipes of the main wastewater treatment plants in La Ribera and Los Alcázares (in this case through the Albujon watercourse) (Fig. 5a).

As a consequence of changes in nutrient input regime, the water column in the lagoon has changed from being moderately oligotrophic to be relatively eutrophic providing conditions for growth of larger phytoplankton cells and subsequent changes in the trophic structure. So, phytoplankton assemblages in 1988 were mainly characterised by small flagellates (Crytomonads, Rhodomons and Pyramimonads), small diatoms (*Cyclotella* spp.) and dinoflagellates (such as *Ceratium furca*, which shows frequent blooms, or *Prorocentrum micans*) (Ros & Miracle, 1984a, b; Gilabert, 1992) with only some short blooms of larger diatoms such as *Chaetoceros* sp. However, in 1997, high densities of bigger diatoms such as *Coscinodiscus* spp. and *Asterionella* spp. were also found.

The two main species of jellyfish (*Rhyzostoma pulmo* and *Cotylorhiza tuberculata*) reached annual mean densities of 0.45 and 2 individuals per 100 m^3, respectively, in 1997, with local maximum densities of the second species that reached 40 individuals 100 m^{-3} and a total population estimated at 46.98 million of individuals (Pérez-Ruzafa et al., 1997, technical report).

A type biomass spectrum with the corresponding taxonomic groups included is shown in Figure 6. By using image analysis (a non-destructive technique), taxonomic information could be linked to individual volumetric measurements, so that each point in the size spectra can also be taxonomically characterised. The spectrum on the Figure 6 ranks the organisms from small flagellates (2 μm diameter) to adult jellyfishes (up to 40 cm diameter). The fig-

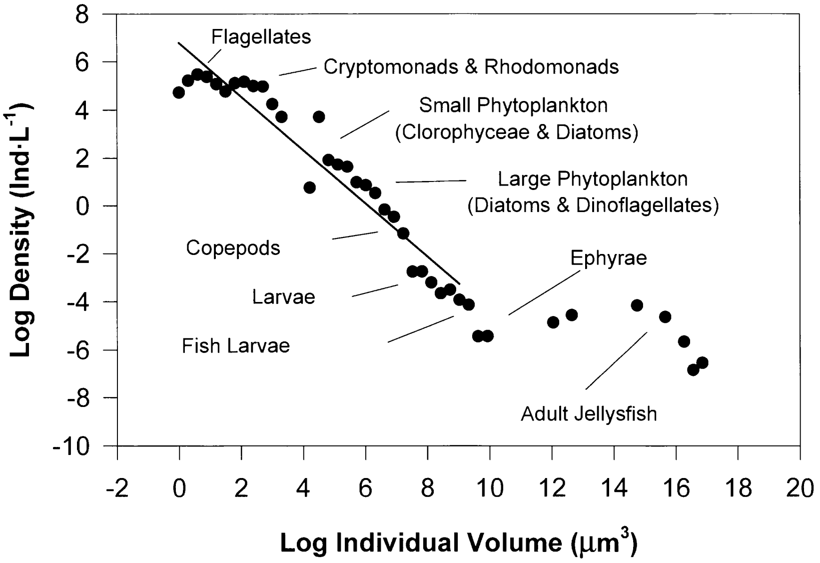

Figure 6. An example of biomass size-spectrum ranking from small flagellates (2 μm diameter) to adult jellyfishes (up to 40 cm diameter). Main large taxonomic groups are indicated on plot each one spanning for several size classes. The regression line is traced excluding the region of gelatinous zooplankton to indicate that only the size range between 2 and 1000 μm diameter was used in the interannual comparison.

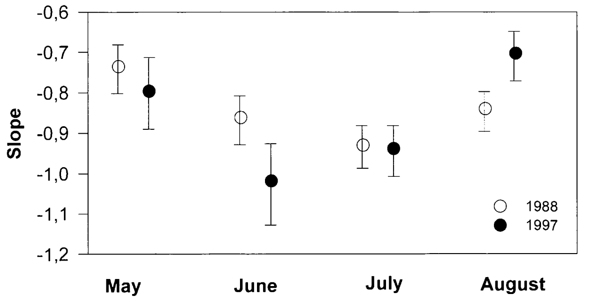

Figure 7. Comparison of the slope values of the biomass size-spectra regression lines between 1988 (open circles) and 1997 (closed circles) in 4 days from May to August.

ure shows a broad range of organisms, including small heterotrophic flagellates, other flagellates such as Cryptomonads and Rhodomonads, small and large phytoplankton, copepods, invertebrate larvae (mostly from benthic organisms) and fish larvae. Ephyrae and adult jellyfishes are included in the right side of the spectra. The regression line excludes the region of gelatinous zooplankton so that only the size range between 2 and 1000 μm equivalent spherical diameter (ESD) was used for the comparisons between 1988 and 1997 (due to lack of jellyfish in 1988). Less negative slopes of regression lines of biomass spectra are found when organisms are accumulated to the right side of the spectra. On the contrary, more negative slopes can be expected when small organisms dominate regardless the total biovolume for the whole range of size classes considered. Although the parameters of the regression line can vary widely depending on the density of individuals in each size class, the slope of the spectra followed a similar seasonal trend in both years (see Fig. 7), suggesting that this trend is imposed by physical factors rather than strictly biological control mechanisms in this particular ecosystem. Figure 8 shows the comparison between spectra showing that total biovolume considered under the above size range was paradoxically always lower in 1997 – with higher nitrate loads, lower concentration of phosphate and very high densities of jellyfishes – than in 1988 – with lower nitrate levels, higher phosphate concentrations and where jellyfishes were not found at the sampling station.

Figure 9 shows the temporal abundance of the jellyfish species during 1997. Each data point on the plot represents the mean density value estimated from all of the 20 sampling stations. *Rhyzostoma pulmo* started to increase in May, while *Cotylorhiza tuberculata* peaked in abundance in June and July, reaching more than 12 individuals per 100 m^3. The total population of jellyfishes estimated in the lagoon by mid summer in 1997 was in the order of 40 million. The gut contents of both species are expressed as percentages in Figure 10. These results show that both species of jellyfish fed predominantly on large diatoms (62.4%) (mainly *Asterionella* spp. and *Coscinodiscus* spp.),

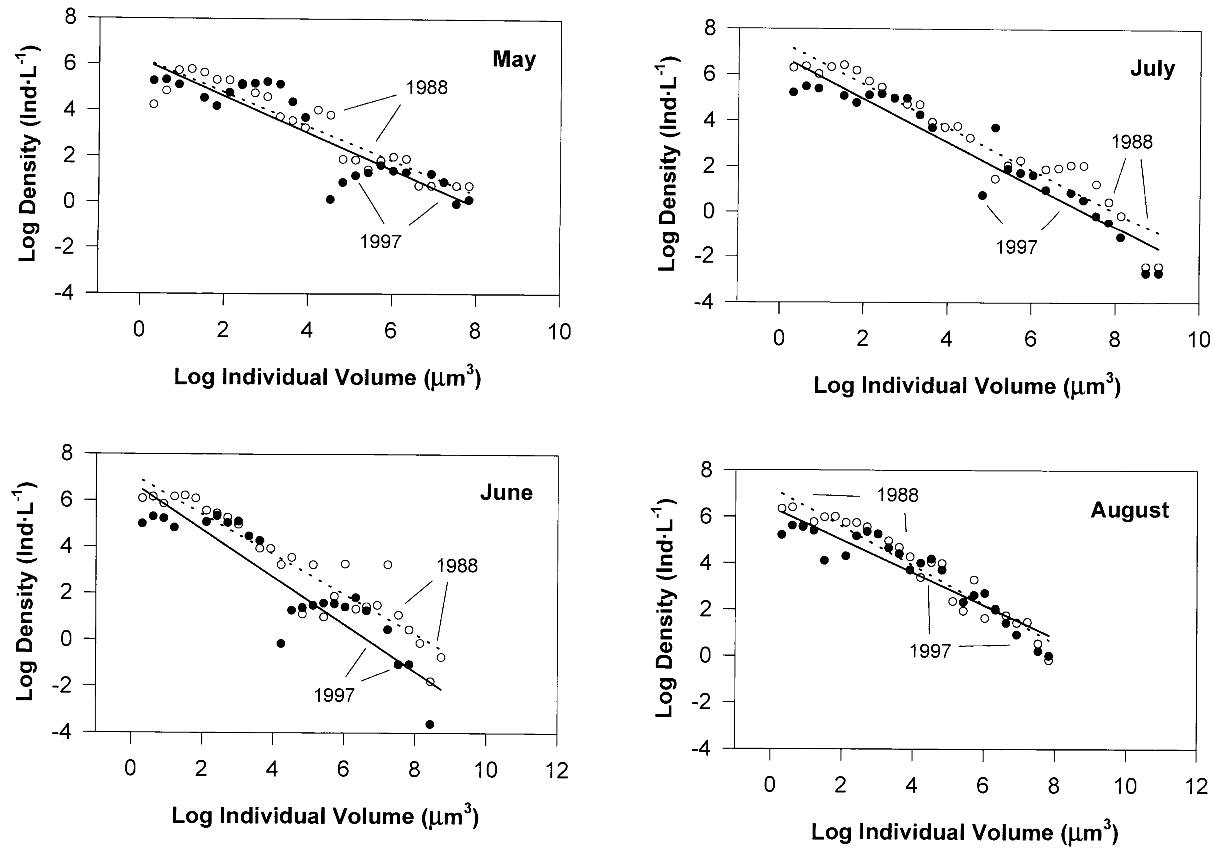

Figure 8. Comparison of biomass size-spectra between 1988 (open circles, dotted regression line) and 1997 (closed circles, continuous regression line) in 4 days from May to August.

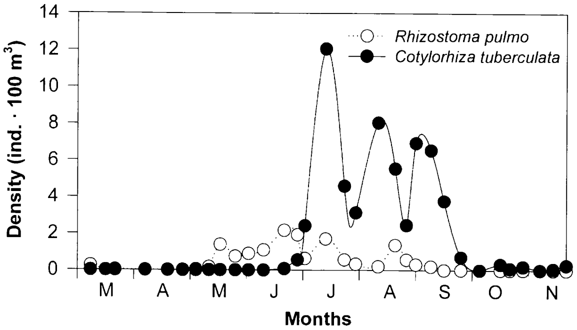

Figure 9. Temporal variation in the mean density of the jellyfishes *Rhizostoma pulmo* and *Cotylorhiza tuberculata* from March to November 1997.

followed by tintinnids. Veliger larvae accounted for 1.8 and 6% while contribution of copepods was 0.5 and 3.17%, respectively.

Discussion

The comparison between nutrient concentrations in the years sampled illustrated changes in the nutrient inputs into the lagoon from 1988 to 1997. In 1988, nitrate seemed to be the main limiting factor for phytoplankton growth. Concentrations were low through the year, contrasting to the higher phosphate values. In 1997, the N:P relationship changed drastically as a consequence of higher nitrate load and phosphate removal. Phosphate patterns in both years showed a strong relationship with seasonal changes in water temperature. The two main reasons for this relationship include: (a) the effect of seasonal increase in the human population when no urban wastewater treatment plants were in operation, and (b) the release of phosphate from sediment (Nixon et al., 1980). Peak tourist season occurs in summer months, but spans between spring and fall with a high increase in population on the lagoon coast. This meant that temperature and phosphate levels peaked together, particularly in

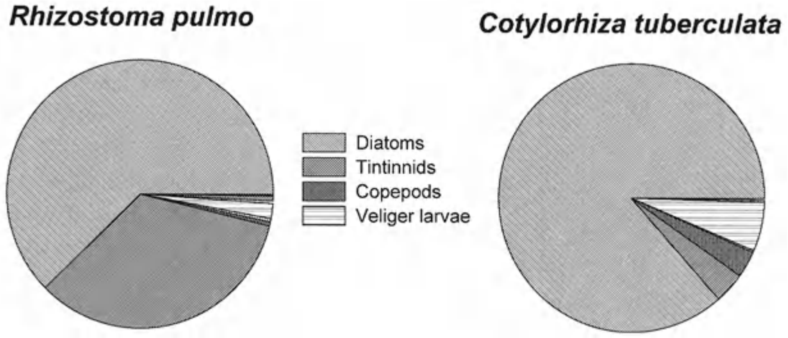

Figure 10. Relative contribution (%) of the main dietary components in the gut contents of jellyfishes *Rhizostoma pulmo* and *Cotylorhiza tuberculata*.

1988 when there was no treated wastewater. In contrast, in 1997, when treatment plants were operational, the relationship between phosphate and temperature was probably due to substrate release rather than wastewater, although a small amount of not treated water could also entered into the lagoon. The situation for nitrate was slightly different. There were no functional watercourses draining into the lagoon in 1988 except during episodic storm and rainfall events. There was a good correlation between nitrate concentration and rainfall values. In contrast, after the rise of the phreatic levels, the major watercourse (El Albujón) was functional in 1997, discharging some freshwater into the lagoon. Spatial distribution of nitrates in the lagoon waters shows a close relationships with watercourse mouths, and mainly with the El Albujon (see Fig. 4). The extent to which this nitrate is diluted in the lagoon depends greatly on water currents, mainly driven by winds. In summary, this results suggest that nitrate was entering into the lagoon via flood runoff in 1988 and through a continuous flow from the main watercourses in 1997. They also indicate that nitrate could have been the main limiting factor for phytoplankton production in 1988, but not in 1997.

According to ecophysiological theory, based on the size dependence nutrient uptake kinetics (Malone, 1980; Hein et al., 1995) and diffusion limitation of nutrient transport (Chisholm, 1992; Thingstad & Rassoulzadegan, 1999), the chance for small cells to grow increases where high nutrient loads provide environmental conditions favouring the shift from small celled phytoplankton to larger cells assemblages. Small phytoplankton should outcompete large phytoplankton when nutrient are scarce, while larger phytoplankton should outcompete small phytoplankton when nutrient level increases. However, competitive success is not only determined by nutrient concentrations. Light or predation avoidance mechanisms can play also a critical role. While small cells are very efficient at absorbing light, larger cells can more easily adapt their pigment contents by packaging them in the chloroplasts improving thus their efficiency. Furthermore, large cells also develop morphological structures, and some of them migratory behaviour, for predator avoidance.

Changes in phytoplankton community structure of the Mar Menor lagoon between 1988 and 1997 are in keeping with the theory, as large sized phytoplankton, such as *Asterionella* or *Coscinodiscus*, were found in 1997 (with higher nitrate levels), but not in 1988 (with lower nitrate levels). However, it is noteworthy that in spite of higher nutrient loads, no very large interannual differences have been observed in the smaller phytoplankton fractions (Gutiérrez, pers.com.). This similarity suggests that both the detritus pathway (Newell, 1982, 1984) and the microbial loop (Azam et al., 1983), characteristic of shallow oligotrophic systems, still play an important role in transferring energy to larger organisms.

It seems evident that presence of large-celled phytoplankton in 1997 in response to elevated nitrate concentrations must have significant implications for the structure and function of the whole planktonic food web, which, should be revealed by the size structure analysis. The spectra slope has been related with the energy flow through the planktonic food web (Silvert & Platt, 1980; Rodríguez et al., 1987; Gaedke, 1993) and with the trophic state of the ecosystem (Sprules & Munawar, 1986). The interannual comparison of four spectra (2–1000 μm ESD) slope in the Mar Menor lagoon did not shows significant differences (p=0.739) indicating that the slope's seasonal trend was maintained during both years showing no dependence on the nutrient load. Furthermore, comparison between spectra revealed that the total biovolume of the size range considered was always lower in 1997 than in 1988 (Fig. 8). This comparison raises the question why, at higher nutrient concentrations in 1997, planktonic biovolume in the studied size range remained lower than at lower nutrient concentration in 1988. The key to solve this paradox may to be in the top-down control mechanisms on the food web structure. Our results support the hypothesis that larger zooplankton, in this case jellyfish, can be an efficient agent top-down controlling the consequences of an eutrophication processes.

In systems where nutrient are scarce, fast-growing small cells can provide available food for certain size range of grazers, resulting in relatively high densities of copepods. Larger cells which grow at higher concentrations of nutrients, may become too large for copepods to graze upon, thereby becoming none available to larger zooplankton (mainly fish larvae and jellyfish). Jellyfish gut contents indicate clearly their preference for large diatoms, tintinnids, veliger larvae and copepods (Fig. 10). High removal rates of larger plankton are expected in the Mar Menor lagoon due to the large number of jellyfishes and their size-selective diet. While the origin of large diatoms in the water column can be explained as a direct consequence of nitrate loads, abundance of veliger larvae is due to the high populations of benthic filter feeders molluscs in the lagoon, mainly *Cardium* and *Ostrea*. Tintinnids, the second most numerically important of gut contents, feed mainly on bacteria, heterotrophic flagellates and small phytoplankton cells (Heinbokel & Beers, 1979). This highlights the importance of the food chains based on small particles. The occurrence of tintinnids and large diatoms in the gut contents of jellyfish supports the notion that an oligotrophic food chain based on small particles and a meso-eutrophic food chain based on large phytoplankton cells co-exist in the Mar Menor lagoon. The effect of jellyfishes removing tintinnids can be, therefore, also seen as an indirect top-down control mechanism on small size fractions. By grazing copepods, jellyfish also act indirectly on small phytoplankton, reducing the top-down control exerted by copepods on small phytoplankton. This finding goes against the traditional view that eutrophication drives systems from small to large cells only. Trade-offs between direct and indirect effects can explain why some eutrophied systems support viable populations of small celled phytoplankton and large populations of large gelatinous zooplankton.

Two main reasons led us to conclude that fishes were not the main top-down controller agent in the Mar Menor lagoon: firstly, most species of fish in the lagoon either live on the seabed or are benthic or benthic feeders (A. Pérez-Ruzafa, 1989). Secondly, the pelagic fishes entering into the lagoon from the Mediterranean (mainly engraulids and atherinids) have drastically reduced their numbers in the Mar Menor lagoon after the enlargement of El Estacio.

So, the results obtained from the Mar Menor lagoon support the conclusion that large gelatinous zooplankton (jellyfishes) exert a strong top-down control on the food web by selective grazing on large diatoms, ciliates, veliger larvae and copepods. Removing large diatoms has a direct effect on nutrients load as they uptake inorganic nutrients from water column, but the simultaneous removal of grazers such as ciliates and copepods reduces the predation pressure on smaller phytoplankton allowing them to flourish. Thus, the feeding preference of jellyfish imposes a combination of direct and indirect effects on the planktonic structure at different size levels. The trade-off between competition for available resources (bottom-up) and predation (top-down) control mechanisms (Lehman, 1991; Cottingham, 1999) results in a planktonic size structure different to that thought to occur under eutrophied conditions. Further studies on trophic control mechanisms could provide very valuable insights for coastal management purposes.

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Some ecological properties in relation to eutrophication in the Baltic Sea

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Abstract

The current published information of the influence of eutrophication on the Baltic Sea is reviewed and summarized. Harmful effects at different levels of the ecosystem are identified, and the spatial and temporal variability of these properties characterized. The Baltic Marine Environment Bibliography was searched on the web, and some 1170 references with 'eutrophication' as a keyword were extracted and analyzed. The most studied regions were the Gulf of Finland (including the Archipelago Sea), Kattegat and the Bothnian Sea. The search was further divided into several parameters (transparency, oxygen/hypoxia, nutrients, primary production/ chlorophyll *a*, algal mats, macroalgae, zoobenthos and fish) related to eutrophication. In most regions, chlorophyll, zoobenthos and fish were the most commonly studied biological and ecological parameters. The linking of eutrophication, ecology and a potential decision-support system is discussed, and related to similar attempts elsewhere.

Introduction

Increased input of nutrients to the Baltic Sea has altered biological structure and ecological processes in both coastal and open-sea areas. By reducing levels of eutrophication, sustainable development of the marine ecosystem may be achieved. As both structure and function of these systems vary greatly along the Baltic gradients of topography, climate and hydrography, we need detailed ecological knowledge of the responses to (reduced) nutrient loading (Gren et al., 1997; Elmgren & Larsson, 2001). Also, public awareness is a vital component when counteracting the negative effects on a Baltic scale, including the drainage area and its population (Partanen-Hertell et al., 1999). Much research on eutrophication is focused on specific ecological processes and properties of local or regional water bodies. The research program MARE ("Marine Research on Eutrophication – A Scientific Base for Cost-effective Measures for the Baltic Sea" – http://www.mare.su.se) aims at analyzing and modeling the negative effects of eutrophication, and at suggesting cost-effective measures for restoration of specific or large-scale water bodies. The goal is a decision-support system for the management of nutrient emissions to the Baltic Sea.

The basic components to be considered when planning measures to control large-scale eutrophication of the Baltic Sea are: (1) the nutrient load to the sub-systems, (2) coastal nutrient budgets and basin-wide models that can be linked to (3) quantifiable ecological changes (structural or functional). These changes are coupled to (4) costs and/or benefits to society, and balanced against (5) costs for effective nutrient reductions. The nutrient reductions, in turn, are coupled to the nutrient loads (1), and hence eutrophication management most effectively be deled with using linked models for ecology, economy, management and policy making (Gren et al., 2000). In this case, 'ecology' must also include information on biogeochemical processes, physical (oceanographic) transports, and nutrient dynamics.

In this paper, we evaluate the status of the Baltic sub-regions in relation to eutrophication, based on information found in reports, publications and on the internet. The aim is to identify gaps in the current knowledge, and to provide an overview of the consequences of eutrophication on regional to basin-wide scales.

A search of available references in the comprehensive Baltic Marine Bibliography (HELCOM) 1970 – to date (http://otatrip.hut.fi/vtt/baltic/intro.html)

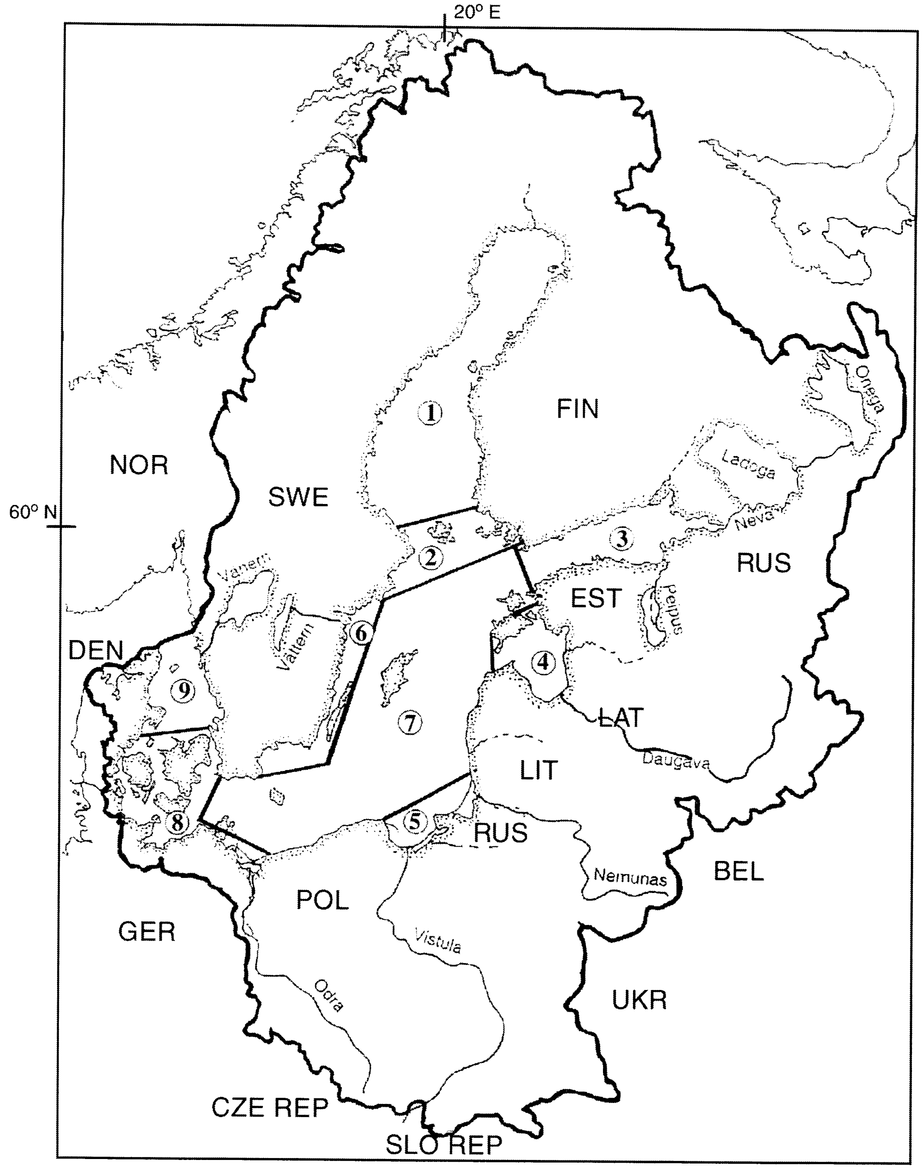

Figure 1. The Baltic Sea drainage area. The sea is divided in the following nine zones: (1) Gulf of Bothnia, (2) Archipelago region, (3) Gulf of Finland, (4) Gulf of Riga, (5) Gulf of Gdansk, (6) Swedish East-coast, (7) Central Baltic, (8) Belt Sea region, and (9) Kattegat. (For more cartographic information on the Baltic Sea and its drainage area, see http://www.grida.no.)

gave no less than 1170 responses on the key word 'eutrophication' alone and in combination with selected parameters (see below).

The Baltic Sea

The Baltic Sea is situated between 54° and 66° N, and 10° 30′–31° E, and is one of the largest brackish water bodies on earth (Fig. 1). The sea is characterized by steep gradients in topography, climate and hydrography, with permanent stratification of the water body over large areas as one key feature. The fjord-like functioning, with sub-basins and sills, further increase the variability of the system (Rumohr et al., 1996). Some basic data are given in Table 1. The brackish character is due to the very narrow link to the North Sea and the inflows of many rivers. The surface salinity is relatively stable, decreasing from 15–25 psu in the Kattegat to 2–3 psu in the northern Bothnian Bay

Table 1. Some basic data of the Baltic Sea area, including Kattegat (Jansson & Velner, 1995)

Parameter	Measure
Area	415 000 km^2
Length	1300 km
Width (W-E)	1200 km
Average depth	60 m
Maximum depth	459 m
Sill depth	17 m
Volume	21 700 km^3
Residence time of water	25 yr.
Total drainage area	1 641 650 km^2

(Elmgren & Larsson, 2001). The critical salinity limit for many marine organisms lies between 5 and 6.5 psu (Kukk et al., 1997), which means that the entire Baltic Sea is a species-poor system, with limited and vulnerable ecological functions (Bonsdorff & Pearson, 1999). The rivers Neva, Narva (Lake Peipus), Vistula, Daugava, Nemunas and Odra (in German Oder) supply most of the fresh water, and over 50% of the total nutrient (P and N) influx to the Baltic Sea. Further, the five largest lakes in Europe – Ladoga, Onega, Vänern, Peipus and Vättern – are all located within the drainage area of the Baltic Sea area (Stålnacke, 1996). About 16 million people live on the coast and there are totally ~80 million inhabitants within the whole catchment area, with severe anthropogenic stress for the marine ecosystem as a consequence. The Helsinki Commission for the Protection of the Marine Environment of the Baltic Sea (HELCOM) has issued a list over sources of special threats – hot spots – in the Baltic Sea drainage area. Over 100 hot spots are defined, mainly industries, municipal wastes and agricultural runoffs (http://www.helcom.fi/). Many of these hot spots relate to eutrophication (municipal effluents, agriculture, aquaculture etc). About one third of the spots are linked to discharges via Poland. The basic characteristics are broadly covered in Wulff et al. (2001).

Nutrient loading

Table 2. Sources of N and P waterborne loads to the Baltic Sea (1000 t y^{-1}), after Gren et al. (1997)

Country	N y^{-1}	P y^{-1}
Poland	312.7	18.4
Sweden	113.7	2.8
Germany	111.8	2.5
Latvia	99.1	1.4
Russia	96.4	3.4
Lithuania	88.9	1.1
Finland	87.1	3.7
Denmark	78.8	1.7
Estonia	37.0	1.2
Distant air deposition	70.5	
Total	**1096**	**36.2**

The nutrient concentrations and the deposition of organic matter have increased considerably in the Baltic Sea during the last century. According to Larsson et al. (1985), the total nitrogen load has increased 4 times and the total phosphorus load 8 times since the beginning of the 20th century. In a more recent publication, it is estimated that 1 096 000 tons nitrogen and 36 200 tons phosphorous (discharges per country: Table 2) are annually discharged to the Baltic (Gren et al., 1997). The trends in nutrient concentrations in the central Baltic Sea during the period 1970–1995 show that the pools of PO_4 and NO_3 have increased significantly both in surface (0–10 m) and deep (100 m) waters, while the concentration of silica (SiO_4) significantly has decreased and that of ammonia (NH_4) remained more or less constant, with a larger annual amplitude (HELCOM, 1996). According to HELCOM (2001) eutrophication is still a major problem, with no real signs of reduced concentrations of nutrients during the latter half of the 1990s. The decreasing silica pool is a phenomenon especially obvious in the Southern Baltic. When the annual diatom blooms diminish, toxic dinoflagellates take over. The reason for this development is both the damming of rivers and eutrophication. Dams convert a river to a lake that increases the residence time of water and improves the light conditions. Thus, the algae, including diatoms, are favored and the sedimentation of Si increases (Humborg et al., 2000). These macro-trends are also reflected on local scales (Bonsdorff et al., 1997a, b). The semi-enclosed situation of the Baltic Sea, and its strong permanent water stratification, long water residence time and low annual mean temperature cause slow mineralization of organic wastes, making the Baltic Sea very vulnerable to anthropogenic influences (Jansson, 1980; Wulff et al., 1990). The nutrient reductions are slow and the effects may last for decades, partly due to long stagnation periods with large hypoxic/anoxic areas and release of hydrogen sulphide (Unverzagt, 2001). A comprehensive review of the present knowledge of these issues is presented in Wulff et al. (2001), and comparative information about the adjacent sea area, the Skagerrak, is presented in Karlson et al. (2001).

Some effects of eutrophication

Coastal areas have naturally higher nutrient values than the open sea, due to inflows from land, more shallow and complex topography, higher water temperatures and restricted water exchange. An archipelago area can serve as a nutrient sink, however. Only a part of all the nutrient input to an archipelago area will reach the open Baltic Sea, and it can be said that the archipelago works as a sewage treatment plant for the open sea (Elmgren & Larsson, 1997, 2001). Increased nutrient concentrations in the coastal areas have increased the amount of particles in the water and thus reduced the transparency. Thus, the Secchi depth has declined from a mean value of 9 m off Hangö in the Gulf of Finland in 1914–1939 to 5 m in the beginning of the 1990s (Jumppanen & Mattila, 1994). The largest eutrophication-related problems in coastal areas are the excessive increases of filamentous algae, disturbing and deteriorating more slowly growing macroalgae and suffocating benthic assemblages (Norkko, 1997). As an example, in the Puck Bay in the Gulf of Gdansk, *Fucus vesiculosus, Furcellaria lumbricalis* and *Chorda filum* have disappeared since the 1970s, and filamentous species of *Ectocarpus* and *Pilayella* are now dominating the area (Ciszewski et al., 1992). The decomposition of these algae may cause further problems, as they can form loose-lying, drifting mats on the bottoms, acting as a physical cover and reducing the oxygen conditions below (Norkko, 1997; Vahteri et al., 2000).

In the open sea, it is harder to distinguish between changes caused by natural processes, such as climatic change, or by pollution and eutrophication (Matthäus & Schinke, 1999; Hänninen et al., 2000). Few long-term biological changes in the pelagic zone have been linked to eutrophication. This does not mean that they are absent; rather that time series are lacking and adequate sampling frequency is seldom achieved (Cederwall & Elmgren, 1990). Primary production in the summer has approximately doubled in the southern Baltic proper since the 1970s, and chlorophyll *a* concentrations have also increased with the same order of magnitude (Anon., 1990; Cederwall & Elmgren, 1990). Mass occurrences of phytoplankton cause an increased enrichment also in the sediments. Beyond the increased biomass, the species diversity and composition of the phytoplankton community will also change (Skei et al., 2000). The three dominating cyanobacteria species in the Baltic Sea are *Nodularia spumigena*, *Aphanizomenon* spp. and *Anabaena* spp. They are all common especially in the Baltic proper and may form dense 'blooms' in July and August. It is suggested that the water stratification with strong halocline and the lack of a thermocline favor the development of cyanobacteria in the Baltic (http://www.mare.su.se). *Nodularia* may be toxic throughout its life cycle but *Aphanizomenon* is non-toxic. The toxicity of *Anabaena* in the Baltic Sea is not clear. As they are able to fix nitrogen, 15–25% of the primary production in the Baltic Sea is made up of cyanobacteria (http://www.mare.su.se). It can be assumed that approximately 180 000–430 000 tons of nitrogen are fixed in the upper mixed layer of the Baltic Sea proper during annual blooms (Larsson et al, 2001).

Increased oxygen consumption in the bottom water appears when excess organic matter produces with the nutrients sink and are decomposed at or near the bottom. This may cause temporary or permanent anoxic conditions with formation of hydrogen sulphide and nutrient flux from the sediment as a consequence (Jansson, 1980; Larsson et al., 1985). The stagnant water in the deep basins enhance the oxygen depletion (Jansson, 1980) and the area of the dead bottoms varies. Currently, the anoxic area is 100 000 km^2 (~25% of the total area of the Baltic Sea), concentrated to the Gulf of Finland, the Baltic proper, the Belt Sea and the Kattegat. Large inter-annual variations occur, and the present development is towards a decreased volume of hypoxia, but with more pronounced anoxia and increased amounts of hydrogen sulphide released into the system (Unverzagt, 2001). As azoic and anoxic bottoms spread, the capacity of the Baltic for neutralizing the nitrogen inputs will probably be reduced and eutrophication intensified (Rosenberg et al., 1990). For coastal waters hypoxia and anoxia are classified as major threats to coastal ecosystems also on a global scale (Diaz & Rosenberg, 1995).

Hypoxic and anoxic conditions in the bottom water can destroy whole benthic communities. A functional selection occurs, as some species are more adapted

Table 3. The geographical and thematical distribution of studies on 'eutrophication' in the Baltic Sea. All studies are taken from one major source: http://otatrip.hut.fi/vtt/baltic/intro.html. Total number of studies analyzed were ~1170. The sums do not always match, as numerous reports contain information on more than one sub-area or theme. Explanations: E=eutrophication, s=Secchi, h=hypoxia, n=nutrients (P, N), pp=primary production, chl *a*=chlorophyll *a*, ab=algal blooms, m=macroalgae, da=drift algae, z=zoobenthos, and f=fish

Parameters:	E	E+s	E+h	E+n	E+pp/chl *a*	E+ab	E+m	E+da	E+z	E+f
Area:										
Gulf of Bothnia	96	5	2	23	16	3	3	1	18	38
Archipelago region	119	18	6	36	32	7	4	11	26	39
Gulf of Finland	103	2	3	57	21	8	6	–	5	9
Gulf of Riga	26	3	1	8	10	–	1	–	4	3
Gulf of Gdansk	13	–	–	2	1	–	–	–	–	1
Swedish E-coast	31	–	1	8	4	–	–	–	13	6
Central Baltic	86	3	6	53	19	5	1	–	19	12
Belt Sea region	64	2	3	30	12	1	1	–	11	9
Kattegat	99	4	11	24	16	9	8	–	21	27

to survive extreme oxygen depletion than others (Rumohr et al., 1996; Bonsdorff & Pearson, 1999). The only organisms that stay alive during long hypoxic periods are those that have the physiological ability to withstand the new environmental conditions (Hagerman, 1998). Their relative immobility prevents most benthic animals from escaping (Baden et al., 1990; Hagerman, 1998). About one third of the bottoms in the central Baltic proper were laminated and devoid of life in the late 1980s, according to Skei et al. (2000). For the fish stocks, better feeding conditions have resulted in increased catches since the 1970s (Nehring, 1991). ICES has measured increased catches of herring, sprat and cod from 450 000 tons in 1966 to 900 000 tons in 1980 and then remaining at that high level (Nehring, 1991). Schematic pathways of the eutrophication processes of the Baltic Sea subsystems are illustrated in e.g. Bonsdorff et al. (1997b) and at http://www.mare.su.se.

The regional coverage of present knowledge

We can conclude that the consequences of the eutrophication process, starting by an increased availability of plant nutrients, have different patterns in different regions and biotopes of the Baltic Sea. The sea can be divided into a series of basins of varying depths, separated by shallow areas or sills. Based upon these criteria, the Baltic Sea is in our study split into nine zones: The (1) Gulf of Bothnia, (2) Archipelago region, (3) Gulf of Finland, (4) Gulf of Riga, (5) Gulf of Gdansk region, (6) East coast of Sweden, (7) Central Baltic, (8) Belt Sea region, and (9) Kattegat (Fig. 1). The changes in structuring and functioning of the ecological system in relation to eutrophication parameters (Secchi depth, oxygen, nutrients, primary production and chlorophyll *a*, harmful algal blooms, macroalgae, drifting algal mats, zoobenthos, and ichthyofauna) are examined (Table 3). Clear similarities and differences between the regions can be seen; changes in nutrient concentrations and in the zoobenthos are common to most of the zones, as are problems with harmful algal blooms. Other parameters seem to vary in significance between regions. Thus, drifting algal mats are an increasing problem in the Archipelago region (Vahteri et al., 2000), while in Kalmarsund at the Swedish East coast, reduced or even locally extinguished fish stocks have worried local residents and perplexed researchers. According to Anon. (1993, 1999) total phosphorus and nitrogen, chlorophyll *a* and Secchi depth are key parameters for interpreting and managing eutrophication. However, various biological end-parameters need to be studied as well. The geographical and thematic distributions of about 1170 reports and references found in the Baltic Marine Bibliography are presented in Table 3. Of this mass of work on Baltic eutrophication since published in 1970, some 640 deal with general eutrophication, 241 include eutrophication+nutrients, 144 include eutrophication and fish, and 131 link eutrophication and primary production/chl *a*. Zoobenthos is coupled to nutrient enrichment in 117 cases. The other parameters are less often directly linked to eutrophication. Among the sub-areas analyzed (see Fig. 1), the Gulf of Finland and the Archipelago Region have received most attention, followed by Kattegat, Swedish coastal waters, and the Gulf of Bothnia. This does not necessarily reflect 'eutrophication', but rather the awareness of the problem (Partanen-Hertell et al., 1999) and the distribution of researchers, and shows the need for joint management on a Baltic scale also in this respect (Gren et al., 2000, Hildén et al., 2000).

There are, naturally, also factors other than nutrient enrichment that contribute to the current situation of the Baltic Sea, such as climatic changes and effects of pollutants. Changes in the meteorological conditions may be of importance, in combination with the anthropogenic effects. Studies on the North Atlantic Oscillation (NAO) show stronger winds and increased runoff when the NAO is high, and an intricate balance between the effects of climate change and anthropogenic factors, such as eutrophication, complicates our interpretation of ecological information (Hänninen et al., 2000; Hoerling et al., 2001). Concerning pollutants, most of the well studied compounds, such as PCB and DDT, show decreasing trends. The same is true for the heavy metals cadmium, copper and mercury, which have been reduced by 50% in the sediment of the Baltic proper since the 1980 (HELCOM, 2001). However, the increased biological particle production due to eutrophication may enhance the export of trace metals into the sediment (HELCOM, 2001), and the links between nutrient enrichment and other pollutants or stresses have attracted interest in recent years (Anon., 2000).

Concluding remarks

As demonstrated in this paper, there is much, but relatively scattered ecological information available on the issue of nutrient over-enrichment (eutrophication) of the Baltic Sea and its sub-basins. Scientists have tried to alert society to increase awareness of these issues (Partanen-Hertell et al., 1999), and to plan for joint-actions to counterbalance the negative effects (Gren et al., 2000). In Cloern (2001) a comparative view of the eutrophication problem is presented on a global scale, and examples from the Baltic Sea are put into a broader perspective. To reach an overall conceptual model, the following aspects need to be accounted for: system-specificity, interactions with other stress-factors (apart from nutrient loading), impacts on a global scale, management, and strategies for restoration and rehabilitation of damaged ecosystems. On a local scale, such a model is used for estuaries both in Europe (Conley et al., 2000) and in the U.S.A. (Bricker et al., 1999), and such a scheme can partly also be applied for the Baltic Sea, and some progress has also been made for predictive purposes (Meeuwig et al., 2000). Today the following statements characterize "the state of the Baltic Sea", and action is being taken to improve the situation:

- no decrease in nutrient concentrations
- increased turbidity & sedimentation rates
- increased growth of pelagic/benthic algae in shallow waters
- coastal waters affected by large amounts of drifting (rotting) filamentous algae
- large bottom areas affected by hypoxia/anoxia (coastal and off shore)
- increased formation of H_2S
- continued reduction/extinction of zoobenthos and disappearance of fish
- extensive harmful (toxic) algal blooms
- altered food webs that provide opportunities for alien species in the Baltic; potential problem
- EUTROPHICATION recognized as "one of the major environmental problems in the Baltic Sea".

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Evaluation of the nutrient inputs to a coastal lagoon: the case of the Ria de Aveiro, Portugal

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Key words: coastal lagoon, plant nutrients, sewage, submarine outfall, eutrophication

Abstract

The Ria de Aveiro estuary-coastal lagoon system of northern Portugal is estimated to currently receive mean annual influxes of total nitrogen (N) and total phosphorus (P) of c. 6118 t y^{-1} and 779 t y^{-1}, respectively, from its influent rivers. In low summer flows the mean N and P fluxes decrease to c. 10% of the annual average. The sewage contribution to the inland-derived N load on an annual basis is c. 5% but, during the summer low flow conditions, the sewage component increases to c. 65% of the total river loading. The sewage contribution to the inland-derived P load on an annual basis is c. 11% but, during the dry season, it is 1.2 times larger than the river-derived flux. The construction of a regional sewer system linked to a submarine outfall, due for completion in 2005, is expected to lead to a reduction in nutrient fluxes from inland to the lagoon of c. 15% for N and c. 26% for P relative to the present values. While this system will reduce the nutrient loading in the upper reaches of the lagoon, an increase in nutrients derived from the ocean is anticipated, due to the proximity of the outfall to the inlet.

Introduction

This paper evaluates the changes in nutrient inputs to the Ria de Aveiro, northern Portugal, resulting from the operation of a new sewer system. This complex, shallow estuary-coastal lagoon system has water quality problems related to organic matter and nutrients discharged from bordering urban and industrial areas (Hall, 1982; Borrego, 1993). A specific problem identified was the organic enrichment of the inner parts of the lagoon. To manage organic enrichment in such a coastal lagoon, it is necessary to control not only the input of organic pollutants but also the input of plant nutrients (Vollenweider, 1992; Harremoes, 1998). This situation in the Ria de Aveiro has led to the construction of a new regional sewer system, with secondary level wastewater treatment, linked to a submarine outfall, due for completion in 2005. The decision to discharge the effluent into the Atlantic Ocean was based on the lower operational costs, compared with full nutrient removal from the wastewater.

In addition to the nutrients associated with wastewater discharged into the lagoon, there are diffuse sources of nutrients from the catchment basins. The contribution of these sources to the total nutrient loading of the system has not previously been quantified, due to a lack of permanent water sampling stations on any of the rivers entering the lagoon. Prior to this study, the only available data were for the upper reaches of the principal influent, the River Vouga, where samples are collected once per month near an urban water supply intake. On the other rivers, only occasional samples have been collected at various flow rates.

The lagoon

Located at approximately 41° N, 9° W, the Ria de Aveiro is the most remarkable geomorphological feature dominating the Atlantic Ocean coast of northern Portugal. It comprises a combined shallow estuary-coastal lagoon with a complex morphology and pro-

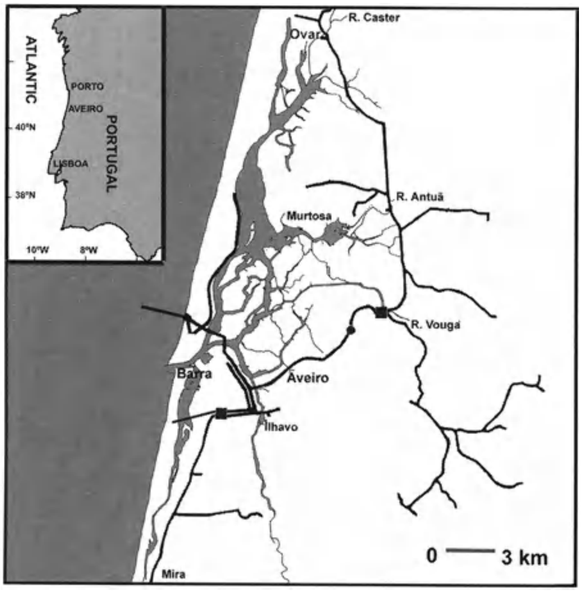

Figure 1. Location map of the Ria de Aveiro, showing channels and towns referred to in the text. The solid black lines indicate the new regional sewer system linked to the submarine outfall.

ductive ecosystem. The physical system is characterised by many branching channels, the most important of which are Ovar, Mira, Ilhavo and Murtosa, that are connected to the ocean by a single tidal channel, via an intervening tidal lagoon (Fig. 1). Water circulation within the ria is driven dominantly by ocean tides transmitted through the c. 4000 m^2 inlet at Barra. The inlet channel is constrained by two converging breakwaters, defining a cross-section some 15 m in depth and 300 m in width. The tide at the inlet is semidiurnal with a maximum range of c. 3 m and tidal wave propagation into the lagoon takes the form of a damped progressive wave (Dias et al., 1999). Whilst the water circulation of the lagoon system is dependent on its connection with the Atlantic Ocean, the system is morphologically unstable, with both the inlet/outlet channel and the lagoon increasing in depth through time. This is largely due to improvements to the inlet/outlet channel, increasing its cross-sectional area, which have led to a steady increase in tidal amplitude (Silva & Duck, 2001). The lagoon itself is very shallow, depths greater than 2 m are restricted to a small fraction of its mean area of c. 66 km^2. The mean water volume of c. 100×10^6 m^3 is close to the tidal prism volume, which presently is c. 83×10^6 m^3 for an average tide. The residence time, based on salinity distribution, is less than 1 day for the central area close to the inlet/outlet channel, and increases to >15 days in the inner areas (Silva, 1994).

The water ebbing from the lagoon forms a plume that was studied by Anderson et al. (1999) and Silva et al. (1999). The observations have shown the complex interaction of the lagoon water with coastal water, transported predominantly southward by a littoral circulation generated by wind and wave action. During the flood tide, it was observed that water flowing from the north rotates to the east and is diverted into the inlet channel. The observed flow pattern suggests that the quality of the water in the littoral zone to the north of the inlet channel will have a significant influence on that of the water entering the lagoon on the flood tide.

The catchment area

The total catchment area of the Ria de Aveiro is c. 3500 km^2 of which c. 2400 km^2 is drained by the River Vouga (Fig. 2). Most of the area drained by the River Vouga is underlain by rocks of low permeability (schist of Cambrian age intruded by Hercynian granite plutons) and is characterised by steep, mountainous slopes. This geological setting, together with the regional weather patterns, is the cause of large seasonal differences between winter runoff events and the summer base flow. Hence, the annual mean runoff is dominated by the extreme events associated with intense rainfall episodes.

During winter and spring the runoff usually ranges from 20 to 60 m^3 s^{-1} but periodic high flow events (>100 m^3 s^{-1}) can cause extensive flooding of the land bordering the inner areas of the lagoon. During summer, the base flow is typically reduced to 3–4 m^3 s^{-1} (Silva, 1994). During these low-flow periods, evaporation exceeds the freshwater input and causes some parts of the lagoon to experience higher salinities (>36 psu) than the local oceanic values. Such conditions result in a detrimental impact on adjacent agricultural fields, which must be protected from saltwater intrusion by the construction of dykes. Thus, it is expected that diverting the freshwater effluent with a maximum flow up to 2 m^3 s^{-1} to the Atlantic Ocean will lead to a more haline aquatic environment for the lagoon ecosystem.

Human presence

The combined population in the total catchment area of the Ria de Aveiro is >600 000, which corresponds to a mean population density of c. 170 inhabitants

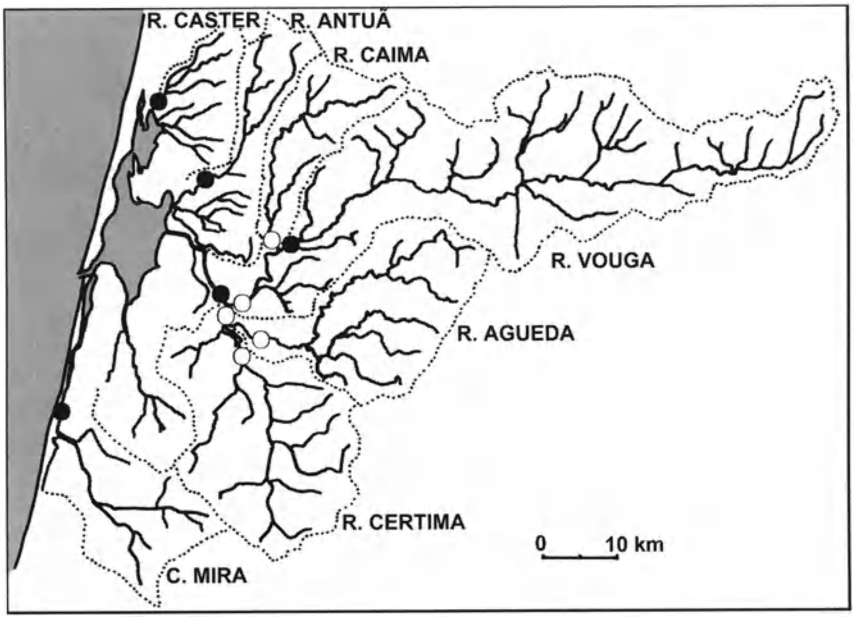

Figure 2. The catchments of the various rivers draining into the Ria de Aveiro, showing principal water sampling stations (solid circles) and secondary sampling stations (open circles).

km^{-2}. The population density is not, however, uniform and varies considerably among the catchment areas of the individual influent rivers. That of the major River Vouga, draining nearly 70% of the total catchment, has a population density of c. 110 inhabitants km^{-2}. To the north side of the lagoon the catchments of the Rivers Antuã and Caster (Fig. 2) contain a much higher population density of c. 470 inhabitants km^{-2} whereas the land surrounding the lagoon and the catchment areas of the rivers to the south have a density of c. 180 inhabitants km^{-2}. In spite of the high densities, the population is distributed sparsely over much of the territory as the large urban areas of Aveiro and Ilhavo (Fig. 1) contain about 11% of the total.

Most of the population within the Ria de Aveiro catchment still use septic tanks or other small systems for sewage disposal. Sewage drainage systems are restricted to the major urban areas and serve c. 37% of the total population (1991 data). Currently the effluent from c. 90 000 inhabitants is still discharged directly into the lagoon by the towns in six municipalities around the ria. Most of the sewage collected receives either poor or no treatment. During the last decade, the need for improved sewage collection has increased in parallel with rapidly expanding urbanisation. The creation of individual connections is, however, lagging behind the construction of a regional sewage system; thus, large numbers of septic tank systems are still in operation.

The nutrient loads transported by rivers to the lagoon are also associated with agricultural activities. The Portuguese National Agriculture Statistics from 1989, obtained from Instituto Nacional de Estatística, Lisboa, were used for characterising the agriculture and cattle production in the catchment area. In the mountainous region, agriculture is restricted to <20% of the area, while in the lower reaches of the rivers and immediately around the lagoon agriculture uses a larger fraction of the area, mainly for cattle farming. The numbers of cattle, sheep, goats, pigs and poultry that are reared in the catchment area of the ria, compared with the numbers of inhabitants, are an index of the importance of animal production. The numbers of animals farmed correspond to ratios *per* inhabitant of 0.4 for cattle, sheep and goats combined, and 0.3 for pigs. These ratios vary little over the area. In the River Vouga catchment area, there is also a large production of poultry, corresponding to 16 birds *per* inhabitant. To compare the importance of this source with the urban sewage load, the numbers of cattle, sheep, goats, pigs and poultry were converted into inhabitant equivalents using rates of excretion of N and P obtained from Benneton (1986) and Ryser (1994). The conversion was based on the N load, which gives values that are 2–3 times those obtained for P. The values of inhabitant equivalents *per* km^2 range from c. 630 for the River Vouga catchment to c. 860 for the rest of the area.

Field methods

The concentrations of forms of N and P in river waters were measured monthly at points on the Rivers Vouga, Antuã, Caster and Mira canal (Fig. 2). The selected sampling stations are located in the lower reaches of the rivers, above the tidal limit. As the lower reach of the River Vouga receives the drainage from important tributaries (Caima, Agueda and Certima) these were also sampled, as well as the River Vouga waters above the confluence of these tributaries (Fig. 2). This sampling strategy was designed to measure the contributions to the nutrient fluxes from catchment areas with different land use practices. The River Agueda drains an area dominated by woodland while the Certima drains an area with important agricultural and industrial activities.

Water sampling began in April 2000 during a period of high river flow and was repeated until September 2000 when river discharge fell to the low summer situation. The sampling dates (see Fig. 3) were chosen taking into account the stability of meteorological conditions. River flow was measured in the lower reaches of the Rivers Vouga, Antuã and Caster with a Valeport BFM001 flow meter, according to the British Standards Institution (1980).

Water samples were collected for the *in situ* determination of temperature, conductivity and dissolved oxygen and additionally 3 l samples were transported to the laboratory for the determination of BOD, pH, ammonia (NH_3 gas electrode), suspended solids (glass-fibre filter), nitrate and nitrite (reduction in Cd column), organic N (Macro-Kjeldahl) and total P (digestion with H_2SO_4+HNO_3 and colorimetry). These analyses were performed according to methods of Eaton et al. (1995). Samples were stored at low temperature (<6 °C) and the analyses were undertaken in the order indicated above and completed within 7 days of collection.

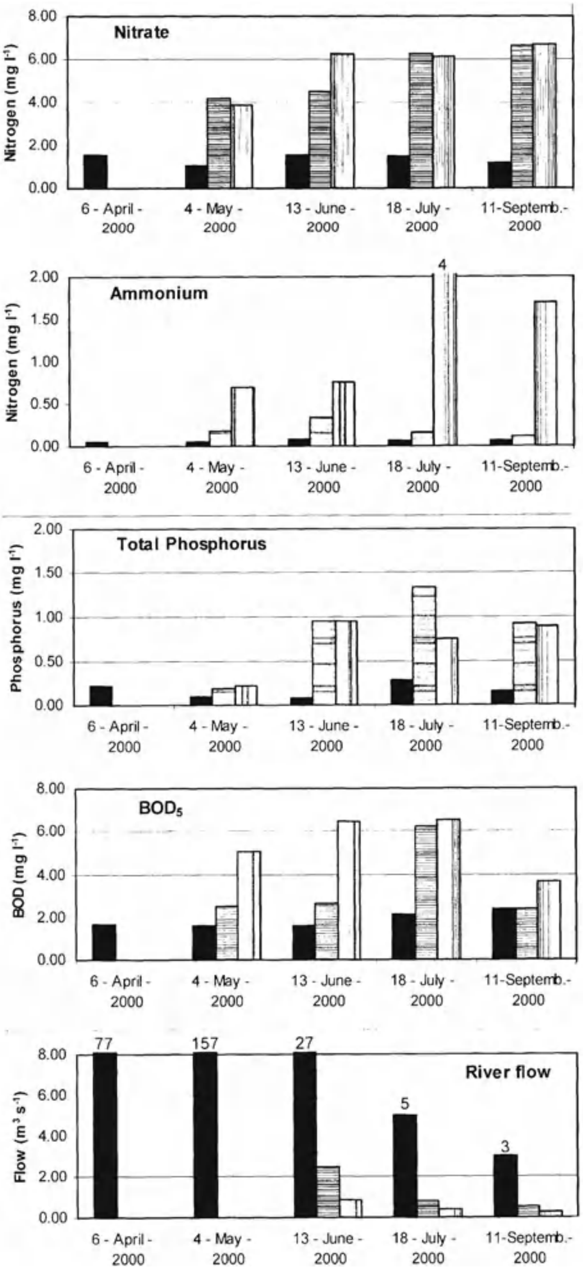

Figure 3. The concentrations of nitrate, ammonium, total phosphorus and BOD$_5$ for water samples collected from the Rivers Vouga (black bars), Antuã (horizontal stripes), Caster (vertical stripes), and the corresponding river flows.

The characteristics of the sanitation systems existing in the towns of Aveiro, Ilhavo and Ovar (Fig. 1) were identified on the basis of the numbers of inhabitants connected and the level of treatment provided to the sewage collected. The existing sewers do not accommodate all the population in the area, are frequently interconnected with storm drainage systems and are connected to poorly operated sewage treatment plants. To obtain an estimate of nutrient fluxes discharged from sanitation systems the following methodology was used:

- The areas covered by sewer systems were identified, based on information from municipal utilities, and the numbers of inhabitants served were estimated, according to the results of the 1991 census.

- Mean wastewater flow into treatment plants, obtained from municipal utilities, was used to calculate the *per capita* effluent loading.
- Concentrations of N and P were measured in samples of effluent collected at various times and dates in Aveiro, Ilhavo and Ovar. The concentrations measured were used to classify the level of treatment provided to the sewage. Defining the level of treatment allows the estimation of the *per capita* load ratings of nutrients in the sewage discharge.

Results

Nutrient fluxes in the sewage

The results of analyses performed on the urban wastewater samples showed that the level of sewage treatment does not remove most of the N and P. The town of Aveiro discharges primary treated effluent. In the larger treatment plant, serving 26 452 inhabitants, the mean total N and P concentrations were, respectively, 60 and 7 mg l^{-1}, but the inflow showed variations indicative of infiltration to the sewers. For a small, well-controlled plant the mean loads, obtained from the utility data are, respectively, 10 and 2 g inhabitant^{-1} d^{-1}. The towns of Ilhavo and Ovar perform a low efficiency secondary treatment. The results obtained for Ilhavo indicate very low loadings of N and P (respectively, 6 and 1 g inhabitant^{-1} d^{-1}) but it was observed that a large proportion of the sewage produced is discharged into the storm water pipes and is therefore not accounted for. The concentrations obtained for the effluent of Ovar were 64 and 17 mg l^{-1}, respectively, for N and P, consistent with a low efficiency treatment. The typical generation rates for total N and total P present in the sewage are, respectively, 11 and 3 g inhabitant^{-1} d^{-1} (Water Pollution Control Federation, 1983; Benneton, 1986). Composition data, obtained from the Aveiro municipal utility, show that for these towns the generation rates are of the order of 10 and 2 g inhabitant^{-1} d^{-1} of N and P, respectively. In view of these values, and taking into account the low level of treatment provided, which will remove only c.10% of the nutrients (Water Pollution Control Federation, 1983), it is estimated that the sewage discharged to the lagoon will correspond to 9 and 2 g inhabitant^{-1} d^{-1} of N and P, respectively.

Nutrient concentrations in river waters

The available data (Fig. 3) show that the major River Vouga is relatively clean: BOD_5 1–2 mg l^{-1}; N-NO_3 1–2 mg l^{-1}; N-NH_3 <0.1 mg l^{-1}; total P <0.2 mg l^{-1}. These values are above those expected for pristine conditions (EEA, 1999), reflecting the agricultural and urban uses in the catchment area which produce a low pollution load. By contrast, the waters of the Rivers Antuã and Caster, draining the areas of highest population density, have characteristics indicative of higher pollution: BOD_5 3–6 mg l^{-1}; N-NO_3 4–6 mg l^{-1}; N-NH_3 >0.2 mg l^{-1}; total P >0.2 mg l^{-1} (Table 1). The two small rivers to the south of the lagoon, though sampled only sporadically, are in an intermediate condition.

The flow measured in the river Vouga (Fig. 3) reached a maximum in May (c. 157 m^3 s^{-1}) and a minimum in September (c. 3 m^3 s^{-1}). However the variation in nutrient concentrations does not show a clear relationship with the flow rate.

Nutrient loading to the lagoon

On the basis of the available information on river discharge (Silva, 1994) and the mean values of the concentrations of total N (nitrate+reduced N) and total P (Table 1), the fluxes of N and P transported into the lagoon are estimated to have mean annual values of 194 g s^{-1} and 24.7 g s^{-1} (Table 2). These river fluxes correspond to annual loads reaching the lagoon of the order of 6118 t for N and 779 t for P. During the low summer, flows the fluxes become reduced to c. 17 g s^{-1} for N and c. 2.2 g s^{-1} for P. According to the number of inhabitants served by sewer systems discharging directly into the lagoon, it is thus calculated that the sewage contribution (9 g s^{-1}) to the total N load is c. 5% of the mean annual river load. For P the sewage contribution (2.1 g s^{-1}) corresponds to c. 9% of the mean annual river load. The large nutrient fluxes in the river runoff also indicate the importance of agriculture-related activities, especially the N load transported into the lagoon, and are in agreement with the higher loads determined for river runoff compared with sewage contribution. However, during the dry summer months, the N contribution from the sewage load to the lagoon (11 g s^{-1}) increases due to recreational use of local beaches (Table 2) and attains 65% of the summer N flux derived from river runoff (17 g s^{-1}). The sewage-derived P flux in the summer months (2.6 g s^{-1}) attains 1.2 times that derived from river runoff (2.2 g s^{-1}).

Table 1. Mean concentrations and standard deviations of nitrate, reduced nitrogen (ammonia and organic nitrogen) and total phosphate, measured in river water samples collected from April to September, 2000

River	N-NO_3 (mg l^{-1})	N-red. (mg l^{-1})	P-PO_4 (mg l^{-1})
Vouga (*N*=5)	1.38±0.24	0.56±0.26	0.17±0.08
Antuã and Caster (*N*=8)	5.56±1.11	1.25±1.14	0.77±0.39
South river (*N*=2)	0.54	1.71	0.65

Table 2. Mean annual and summer river discharges and corresponding nitrogen and phosphorus fluxes to the lagoon of the Ria de Aveiro, compared with sewage discharges to Ilhavo and Ovar channels and contributions to the nitrogen and phosphorus fluxes from sewage discharges

Source	Water discharge		Nitrogen flux		Phosphorus flux	
	Mean annual (m^3 s^{-1})	Summer (m^3 s^{-1})	Mean annual (gs^{-1})	Summer (gs^{-1})	Mean annual (gs^{-1})	Summer (gs^{-1})
River input						
Vouga	50	4	97	8	8.5	0.7
Antuã+Caster	10	1	68	7	7.7	0.8
South rivers	13	1	29	2	8.5	0.7
TOTAL RIVER	**73**	**6**	**194**	**17**	**24.7**	**2.2**
Sewage input						
Ilhavo channel	0.12	0.12	6	6	1.4	1.4
Ovar channel	0.06	0.06	3	3	0.7	0.7
Beaches	–	0.05	–	2	–	0.5
TOTAL SEWAGE	**0.18**	**0.23**	**9**	**11**	**2.1**	**2.6**

Discussion

Regional sewer system

The technical solution adopted to improve the sanitation situation of the municipalities bordering the lagoon of the Ria de Aveiro was to build a regional system. This would intercept all the sewage collectors and transport the waste to central treatment facilities, prior to discharge via the outfall, located 3 km to the north of the ria mouth, into the Atlantic Ocean (Fig. 1). Some major aspects of this regional system are:

- The number of inhabitants served will increase from c. 100 000, at the beginning of 1998, to >280 000, when the system will be completed in 2005.
- In addition to treated sewage, treated industrial effluents (mainly pulp and paper) will also be transported and discharged to the Atlantic Ocean.
- The interception network will comprise 176 km of pipes plus a further 3 km for the submarine outfall.
- The system includes two treatment stations providing secondary level treatment, and 51 pumping stations.
- The investment cost is of the order of 120×10^6 Euro, financed by the European Union.
- The system is being built progressively from the ocean outfall to the two treatment plants and thence to the more remote parts of the catchment area.

Expected changes in the Ria de Aveiro

When all of the connections to the regional sewage system are completed, the sewage from >280 000 people, corresponding to a N flux c. 30 g s^{-1} and a P flux c. 7 g s^{-1}, will ultimately be discharged into the Atlantic Ocean. As a consequence, it is expected that there will be a similar reduction in the annual mean nutrient loading to the lagoon from the catchment area. Reductions in nutrient fluxes to the lagoon of c. 15% for N and c. 26% for P are thus expected relative to the present situation (Table 2). The new collection system will, however, serve only a small proportion (c. 100 000) of the inhabitants whose sewage is currently discharged into the rivers. The other important nutrient sources related to agricultural activities will also remain unchanged. Thus, the runoff loading of N from diffuse sources will be little affected by the new system. However, the P load reduction from point sources will be significant, especially during the summer.

Once the regional sewer system is fully operational, the areal loads, based on the calculated mean annual fluxes, will decrease from the present value of c. 97 g m^{-2} y^{-1} to c. 83 g m^{-2} y^{-1} for N and from c. 13 g m^{-2} y^{-1} to c. 9 g m^{-2} y^{-1} for P. Even with these reduced loads, the lagoon will have a nutrient loading similar to other coastal systems (Vollenweider, 1992) and hence a potential for continued eutrophication. Historical evidence suggests that the Ria de Aveiro has sustained a highly productive ecosystem. For example, over 100 years ago large amounts ($>10^5$ t) of sea grasses and algae were collected annually from the inner areas of the lagoon.

Besides the reduction in mean annual loads, the changes in timing and location of nutrient fluxes must also be taken into account (Lijklema, 1998). During the dry summer months, the expected reductions in the input of sewage to the lagoon will decrease the total fluxes by c. 39% for N and 54% for P. Another expected change concerns the relative importance of nutrients from fresh water and oceanic sources. The nutrients discharged from the new regional sewerage system will potentially contribute to an increase in the fluxes entering the lagoon via the inlet channel. Remote sensing observations have clearly demonstrated that, the littoral circulation occurs predominantly in a southward direction and on the flood tide landward transport of fine particulate materials from the littoral zone into the ria system takes place (Anderson et al., 1999; Silva et al., 1999). This transport process, which was not foreseen prior to the construction of the outfall, has two important effects on the nutrient balance of the ria. First, the water ebbing from the lagoon is transported to the south and the return during the flood tide is reduced. Secondly, the water flooding into the lagoon comes mainly from the north and northwest and so the effluent discharged from new submarine outfall will influence its quality. Thus, in terms of the trophic status of the lagoon, it is anticipated that, although there will be a reduction in the nutrient input from inland, an increase in nutrients derived from the ocean is a possibility that will require monitoring.

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Evaluation of a eutrophic coastal lagoon ecosystem from the study of bottom sediments

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Key words: organic carbon, nitrogen, phosphorus, sediments, eutrophication, coastal lagoon, Northern Adriatic

Abstract

Morphological reconstruction and biogeochemical characterisation of the lagoon of Comacchio (Italy) were carried out in order to provide recommendations for the recovery, conservation and sustainable management of the Fattibello-Spavola coastal lagoon ecosystem. Samples were taken in two seasons: July 1997 and November 1998. The irregular morphology of the Fattibello lagoon affects depositional processes and seawater exchange (tidal currents). Several depressions retain part of the dense water of the saline wedge; these stagnant waters became sinks for fine terrigenous and organic matter. The basin is already extensively supplied with N and P compounds from land. The inflow is demonstrated by the large quantity of organic C, N and organic and inorganic P compounds in the superficial sediment. The ratios between the various forms of macronutrients indicate that the organic matter is primarily of autochthonous origin, with relatively low C/N ratios (8.4±0.6 and 8.1±0.6 in July and November, respectively). Shallow areas were almost always oxygenated by tidal currents and thus rich in organisms, with a predominance of molluscs and *Ficopomatus enigmaticus*. However, the trophic equilibrium of the ecosystem was affected by the accumulation of organic matter in the depressions, favoured by the increased hydrodynamics during the autumn. These accumulations generate high oxygen consumption and release considerable quantities of nutrients into the water column, with the risk of serious dystrophy throughout the basin during the summer. Local dredging and an improvement of the circulation have been suggested and carried out to contain these processes. Reclamation measures in the longer term were proposed.

Introduction

The Fattibello-Spavola coastal lagoons form part of the Comacchio lagoon area, in turn part of the Po Delta Natural Park. The Ramsar Convention on Wetlands (1971) for the conservation, development and use of wetland resources has covered the Comacchio lagoons since 1981. These marshy basins were in the past the site of extensive land reclamation, fishing and salt production activities. More recently, the salt works have been abandoned in place of intensive fish farming. This has only made the basins' hypertrophic conditions worse with the exception of the Fattibello-Spavola lagoons, which began to play a fundamental role in flushing of the entire Comacchio lagoon system.

More recently, disturbance of the trophic balance has extended to the Fattibello and Spavola lagoon area, with hypertrophic conditions and anoxic crises, particularly intense during the summer. This led the local authorities and Emilia-Romagna Region to carry out dredging operations and dig new channels in the Fattibello-Spavola lagoons in order to improve water circulation and oxygenation in association with tidal currents.

As no previous information was available, a research programme was funded to study morphology, sedimentology and geochemistry of the two basins in order to assess the trophic state of the system, better address the dredge operations and examine their effects on the bottom.

Description of site studied

The Fattibello-Spavola lagoons are located near the city of Comacchio (Fig. 1) and cover a total surface area of about 7.3 km^2. Their hydrodynamics are influenced: upstream, by the inflow of freshwater from the Higliarino channel (liquid flow rate: 328 10^6 m^3 y^{-1}; solid flow rate: 7532 tons y^{-1}) and the Fosse channel (liquid flow rate: 53 10^6 m^3 y^{-1}; solid flow rate: 1471 tons y^{-1}), and downstream, by the inflow of sea water, which is regulated by the tidal cycle through the Migliarino channel and the Logonovo channel. The tidal cycle has a considerable influence on water circulation in the lagoons with an excursion between high and low tides of as much as one metre. The hydrodynamics of the lagoons are also influenced by wave motion generated by predominant winds coming from the south east (*scirocco*) or from the north east (*bora*); the latter is extremely intense in winter.

The tributaries flowing into the Fattibello-Spavola lagoons receive waters from the Volano Po River and drainage waters from adjacent reclaimed lands. A number of sub-lagoon channels, with partially emerging irregular banks, provide a direct link in the basins between the freshwater tributaries and the salt water channels. At the same time, a sub-lagoon channel separates the Fattibello from the Spavola lagoon.

Materials and methods

Field activities

Two sampling cruises were carried out, one before the dredging operations (July 1997) and one during digging operations (November 1998) in order to evaluate the dredging improvements on the basin and to assess the seasonal variations in the trophic state of the ecosystem. The bathymetry of both basins was reconstructed in detail by means of an echo sounder during the first sampling. Problems associated with the shallow depth required development of innovative instrumentation and methods (Masini & Gasperini, 1997).

Both the water column and bottom sediments were sampled at 47 stations (Fig. 1) in July, while sediment samples were taken from a smaller number of selected stations in November. Temperature, salinity, oxygen reduction potential and pH were measured in the water column at two or three levels. Bottom sediment was sampled using a box corer to a maximum depth of 17 cm and subsequently sub-sampled at 1–2 cm thick levels with temperature, oxygen reduction potential and pH measurements being taken on site.

Laboratory activities

The samples were examined by optical microscope in order to estimate the biogenic fractions. Grain size analysis involved an initial wet sieving of sediment samples to separate the finest fractions (<63 μm) from the coarser fractions (>63 μm). The finer fractions were subsequently analysed by X-ray sedigraphy. Given the high content of organogenic residue, the >63 μm fraction was further sieved to 500 μm in order to differentiate between a sandy fraction of between 63 μm and 500 μm and a coarser fraction of >500 μm. Mineralogical composition was determined through XRD and the percentages of the most abundant minerals were evaluated following Cook et al. (1975). Total and organic carbon and total nitrogen were determined by means of an CHNS elemental analyser (Froelich, 1980; Hedges & Stern, 1984). The sub-samples for organic carbon were decarbonated using 1 m HCl and dried at 60 °C. The reproducibility was satisfactory with an average relative standard deviation (RSD) for replicate analyses of 0.5% for total carbon, 0.9% for organic carbon and 1.3% for nitrogen. The inorganic carbon content was calculated from the difference between total and organic carbon. Total and inorganic phosphorus extractions were conducted at room temperature and agitated for 16 h with 25 ml of 1M HCl (Aspila, 1976). The total phosphorus sub-sample was first ashed at 550 °C for 2 h. Reactive phosphates in all extracts were determined using the colorimetric technique (Strikland & Parson, 1972; Griffiths, 1973). The reproducibility was satisfactory with an average relative standard deviation (RSD) for replicate analyses of 2.1% for total phosphorus and 3.3% for inorganic phosphorus. The organic phosphorus content was calculated from the difference between total and inorganic phosphorus. The total manganese concentration was determined in a number of samples by strong acid digestion (Miserocchi et al., 2000). Sequential selective extractions were also performed in order to determine the type of bond attaching the phosphorus to the solid phase (Barbanti & Sighinolfi, 1988). Subsequently, all the solutions were analysed via ICP-MS and ICP-AES for Mn and P fractions, respectively.

Multivariate statistical analysis was applied to the data set. A Q-Factor Analysis (Klovan & Imbrie, 1971; Davis, 1973; Klovan & Miesh, 1975) was chosen with $\cos\theta$ as the coefficient of similarity applied to a matrix of normalised data.

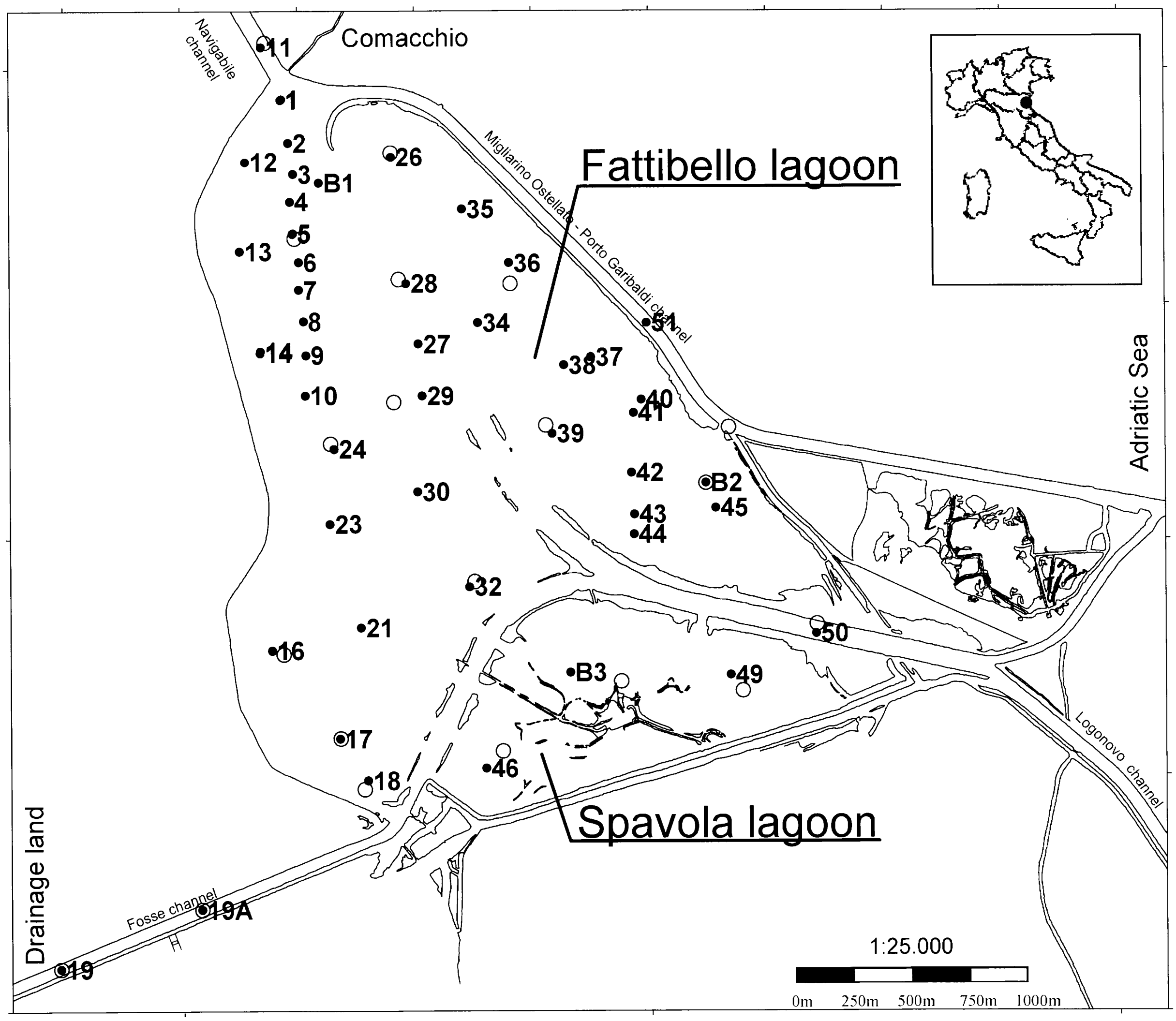

Figure 1. Map of the Fattibello and Spavola lagoons and location of sampling stations in July 1997 (●) and November 1998 (○).

Results and discussion

Morphology

A detailed morphological reconstruction of the basins was carried out during the summer cruise, before the sampling operations. The mean depth of the lagoon basins was about 70 cm, reaching maximums of 2.5–3 m within the sub-lagoon channels. The bottom was extremely uneven (Fig. 2) with sub-circular raised areas and depressions. Near the inflow of the Navigabile channel in the northern zone of the Fattibello, a submerged fan directed southwards was present. With the exception of the sub-lagoon channel, the deepest zone in the Fattibello lagoon lay in the north western sector, immediately to the south of this fan. The Spavola lagoon was everywhere less than 1 m deep.

Texture and composition of sediment

Observations made during the sampling campaign identified zones of the Fattibello lagoon colonised by constructor organisms (*Ficopomatus enigmaticus*) capable of influencing deposition in the lagoon both by biogenic constructions and by bioclast production. Optical microscope observation showed the >500 μm grain size fraction to be composed almost exclusively of bioclasts, consisting of mollusc shell fragments, while the finer fractions (<500 μm) were made up of a mixture of lithoclasts and bioclasts. In terms of grain

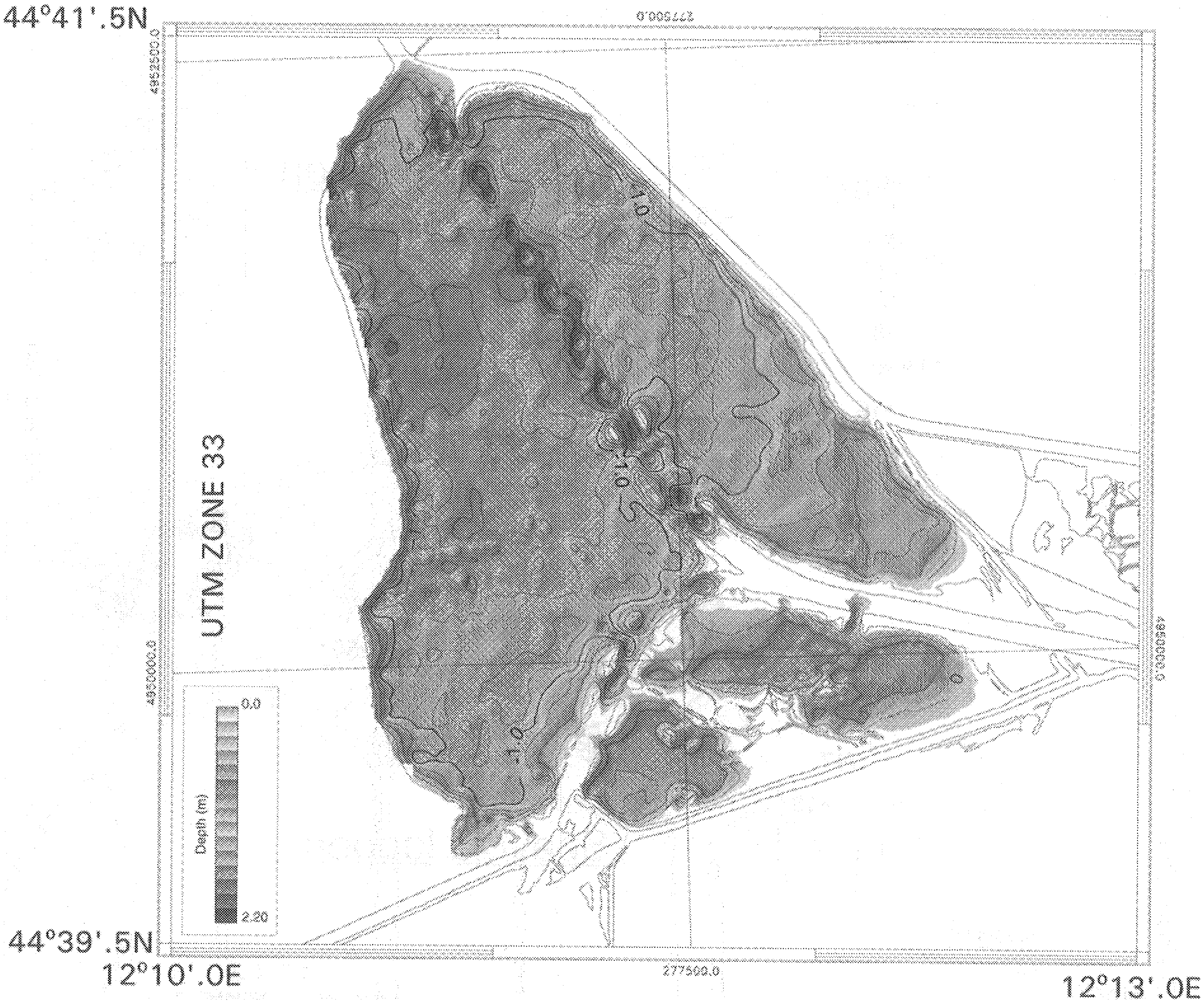

Figure 2. Morpho-bathymetric map (shaded relief) of the Fattibello and Spavola lagoons with 20 cm isobaths.

size, in July, the coarser fraction (>500 μm) of superficial sediment is more abundant in the northern apex and southern part of the basin where it reaches as high as 50%. More modest percentages were observed in the Spavola lagoon and in the sediment of the channels (Table 1). Values tend to diminish at sub-superficial levels. After stormy events, in November there was a mean enrichment of coarse clasts to the detriment of finer fractions in the shallower areas, and in particular on the eastern side, of the Fattibello lagoon. The percentage of the sandy fraction was highest in correspondence to peaks of the >500 μm fraction, with values of more than 15% (Stations 1, 3, 21, 29). The percentage of sand was much lower in the Spavola lagoon and in the channels. The clayey-silty fraction (<63 μm) was concentrated in the internal channel and central eastern zone of the Fattibello lagoon and in the Spavola lagoon with percentages of more than 90% in July. The muddy fraction also characterises the sediment in the bed of the tributary channels upstream of the lagoon area (Stations 19, 19a, 11). The fine sediment fraction tends to increase with sediment depth, from 68.1% ± 22.6 to 70.5% ± 22.3 in Fattibello and from 93.5% ± 5.7 to 96.5% ± 1.7 in Spavola.

Mineralogical analysis identified a carbonaceous component consisting of calcite and dolomite (terrigenous), Mg-calcite (autochthonous *Ficopomatus enigmaticus* shells) and aragonite (mollusc shells) reaching a mean of 50% in superficial samples in Fattibello. Mg-calcite is distributed throughout the Fattibello basin, reaching more than 30% in areas near the colonies of organisms (Stations 1, 26 and 34). Mg-calcite is absent from the Spavola lagoon (Table 1). Aragonite is concentrated in the central and southern band of

Table 1. Average values and standard deviation of sediments (July 1997 and November 1998) and water parameters

		Fattibello Lagoon		Spavola Lagoon		Channels	
		July 1997	November 1998	July 1997	November 1998	July 1997	November 1998
Sediments							
H_2O content	%	63.6 (7.1)	64.8 (8.3)	62.4 (5.8)	62.1 (7.9)	65.3 (13.0)	71.2 (10)
>0.5 mm	%	20.9 (18.0)	24.0 (18.1)	3.5 (3.4)	10.8 (5.8)	2.6 (5.4)	2.6 (5.7)
0.5–0.063 (mm)	%	7.1 (5.0)	7.0 (4.1)	2.3 (0.7)	4.5 (3.5)	19.2 (12.8)	14.2 (15.3)
<0.063 mm	%	72.0 (20.9)	69.0 (21.2)	94.5 3.8)	84.4 (5.9)	78.2 (13.2)	83.2 (19.8)
inorganic C	%	3.72 (1.70)	3.79 (0.99)	2.13 (0.19)	2.35 (0.31)	1.64 (0.31)	1.96 (0.44)
Quartz	%	24 (7)	21 (6)	29 (3)	27 (2)	28 (2)	24 (2)
Plagioclase	%	9 (3)	9 (3)	12 (3)	10 (1)	11 (3)	13 (2)
Feldspar	%	4 (4)	4 (6)	5 (1)	5 (1)	7 (2)	14 (6)
Calcite	%	18 (4)	15 (8)	18 (2)	18 (2)	17 (6)	15 (2)
Dolomite	%	3 (1)	3 (2)	4 (1)	4 (1)	4 (1)	3 (2)
Mg-calcite	%	9 (10)	13 (20)	0 (0)	1 (2)	3 (5)	1 (3)
Aragonite	%	11 (11)	9 (20)	4 (5)	5 (5)	0 (0)	2 (3)
Illite	%	13 (4)	14 (5)	19 (3)	19 (2)	15 (3)	15 (3)
Chlorite	%	3 (1)	(1)	4 (0)	4 (1)	3 (1)	3 (1)
serpentine	%	2 (1)	2 (1)	3 (1)	3 (0)	2 (2)	4 (2)
Eh	mV	−273 (102)	−236 (101)	−264 (66)	−119 (121)	−310 (97)	−317 (46)
pH		7.64 (0.65)	7.85 (0.37)	7.29 (0.15)	7.62 (0.36)	7.12 (0.33)	7.75 (0.34)
organic C	%	2.62 (0.71)	2.68 (0.73)	2.44 (0.52)	2.37 (0.31)	2.34 (1.16)	2.85 (1.04)
total N	%	0.35 (0.10)	0.20 (0.09)	0.31 (0.05)	0.33 (0.04)	0.27 (0.12)	0.41 (0.10)
C/N		8.9 (1.6)	8.4 (1.5)	9.2 (0.9)	8.5 (0.7)	10.2 (0.9)	9.2 (1.10)
total P	$\mu g\ g^{-1}$	837 (264)	795 (162)	692 (173)	654 (93)	1088 (514)	986 (282)
inorganic P	$\mu g\ g^{-1}$	580 (174)	517 (79)	471 (46)	469 (72)	779 (328)	758 (291)
organic P	$\mu g\ g^{-1}$	257 (126)	278 (121)	220 (138)	185 (68)	310 (200)	229 (66)
Waters							
Salinity	PSU	top	12 (3.3)		17 (2.2)		8.7 (9.8)
		bottom	14.7 (4.4)		18.7 (1.4)		16.3 (5.1)
Temperature	°C	top	27.5 (1.1)		28.3 (0.4)		26.2 (0.8)
		bottom	27.2 (0.9)		27.8 (0.3)		25.2 (0.5)
pH		bottom	8.52 (0.6)		7.94 (0.2)		n.d.
Eh	mV	bottom	−4.0 (74.7)		−38.6 (61.0)		n.d.

Fattibello reaching peaks of 40%, while, the content is lower with peaks of 7% in the Spavola lagoon.

A siliceous component is also present in addition to the carbonaceous component, consisting of quartz, plagioclases, feldspars and clayey minerals (illites, chlorites and serpentine).

Quartz, plagioclases, and feldspars are mainly concentrated in the Fattibello lagoon, with the highest values in the northern zone and in the south eastern apex and the lowest values on the eastern side and in the central band of the basin.

Depositional setting

The morphological, grain size and mineralogical data construct a profile of a lagoon system with irregular morphology and depositional processes controlled by the inflow of predominantly fine silicoclastic terrigenous material and by predominantly carbonaceous autochthonous biogenic components (showed by Mg-Ca and Aragonite minerals). The depositional profile provided evidence of silting up (highlighted by detailed morphology; Fig. 2), particularly in Spavola, where the deposition of muddy material is accompanied by poor water circulation. On the other hand, energy conditions are still sufficient to oppose this process in Fattibello. The considerable tidal excursion enables the salt water wedge to penetrate the mouth of the main tributary (Navigabile channel), carrying with it as it withdraws many of the fine sediments in the

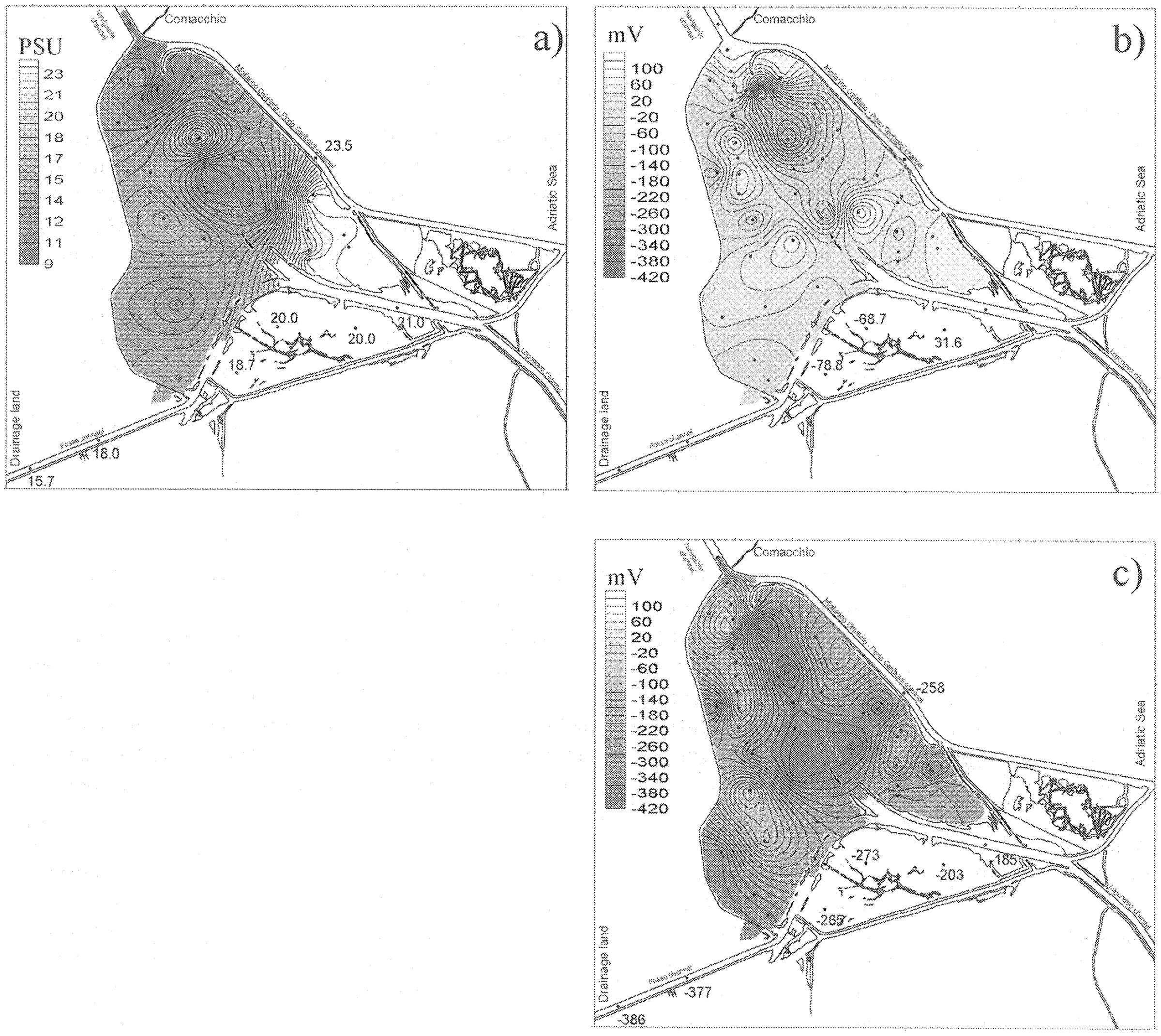

Figure 3. Areal distributions of salinity (a) and Eh (b) of bottom waters and Eh of superficial sediments (c) in July 1997. The bottom water salinity (between 8.6 and 22.7 PSU) reflects the morphological irregularities of the basin. If very shallow marginal zones characterised by direct seawater influx are excluded, the highest values occur at the deepest point of the sub-lagoon channel (St. 28: 19.2 PSU), where salinity and accentuated reducing conditions (lower Eh values measured both in superficial sediment and in bottom water, St. 28: −420 mV and −253 mV, respectively) underline the presence of dense stagnant waters in anoxic conditions in the deeper zone of the lagoon.

basin, sometimes re-suspended by storms. However, the irregular morphology of Fattibello prevents the salt water wedge (Fig. 3a) from withdrawing completely and the morphological depressions retain stagnant still water, becoming a trap for fine inorganic and organic sediment. In the depressions, the pycnoclines are broken and re-suspension takes place only during strong storms.

Physical and chemical properties

Water and bottom sediments physical and chemical properties provide information of great importance in assessing the preferential accumulation sites and autochthonous organic matter transfer processes in a shallow basin.

Eh values measured in the superficial sediments (Fig. 3c, Table 1) indicate a bottom with predominantly reducing conditions both inside the lagoons (Fattibello: −231 mV ± 109; Spavola: −247 mV ±

38) and along the tributary channels (Station 11: −421 mV; Station 19: −386 mV). This situation is locally aggravated also by the bottom water anoxic condition (Fig. 3b, Table 1) and validates the hypothesis of the existence of dense and stagnant water, which persisted in the depressions for long periods as suggested by the depositional profile. Conditions in the sub-superficial sediment are even more reducing. Low Eh conditions persist at the sediment surface in November, but less marked than in July, particularly in the Spavola lagoon (Fattibello: −193 mV ± 110; Spavola: −53mV ± 383). The pH values in the Fattibello lagoon are lowest in the north eastern area, both in superficial and sub-superficial sediment. In general, values tend to decrease with increasing sediment depth (Table 1).

Organic matter

Mean organic C concentrations in July are similar in the superficial sediments throughout the lagoon area (Fattibello: 2.48% ± 0.66; Spavola 2.47% ± 0.29; Fig. 4). The areal distribution is similar in sub-superficial levels, but surprisingly, mean values (2.62% ± 0.72) are higher than in the superficial sediment in the Fattibello lagoon. This suggests that when flow rates are high in tributary channels upstream of the lagoon area, a considerable quantity of sediment is brought into the lagoon, capable of burying large quantities of newly formed and as yet not degraded organic matter.

The mean organic C concentrations in the superficial sediment in Fattibello increase (2.74% ± 0.73) in November and assume a different areal distribution. Values increase also in the channels and in the lowest areas of the basin (Station 28: 2.94% in July and 3.63% in November), in other words, in zones characterised by fine terrigenous deposition and very low Eh values. This confirms and underlines the effect of the accumulation of fine deposits rich in organic matter following autumn storms. Concentration values are slightly lower (2.37% ± 0.3) in the Spavola lagoon, probably as a result of dredging or for seasonal reasons, or both.

Total N concentrations in the superficial sediment vary only slightly between the two seasons (Table 1). The areal distribution is similar to that of organic C, while further down in the sediment, values tend to decrease.

The C/N ratio provide information on the origin of organic matter, where higher ratios indicate terrigenous organic matter and lower ratios autochthonous organic matter (Redfield et al., 1963; Müller & Malthesius, 1999) and its state of decomposition (when organic matter is decomposing, the N is mineralised faster). The C/N ratio is lowest in both seasons on the north eastern side of the Fattibello lagoon and highest in the submerged fan near the mouth of the Navigabile channel. The range of variability is low (Table 1) and individual values are generally such as to suggest that organic matter in the basin is essentially autochthonous (always <10 in superficial sediments). This is associated with a minimum quantity of allochthonous organic matter at the mouth of the Navigabile channel.

The high percentages of organic C and total N and C/N ratio values are indicative of the high level of primary production in the basin and the scarce input of terrigenous organic matter.

The average values of organic C and total N concentrations measured in the lagoon area (Table 1) were higher than those measured in other lagoon basins such as the Sacca di Goro (Frascari et al., 1988) or sea basins such as the North Adriatic (Astorri et al., 1983; Giordani & Angiolini, 1983; Boldrin et al., 1988; Faganeli et al., 1989; Price et al., 1993; Matteucci & Frascari, 1997). A comparison of the results of this study with others carried out in the Baltic Sea (Newuman & Leipe, 1998; Müller, 2000) shows a lower content of organic matter and C/N ratios that confirm the high autochthonous production of the Fattibello-Spavola lagoons.

The mean concentrations of organic P in the sediment are generally high and similar throughout the lagoon system (Fattibello: 257 μg g^{-1} ± 126; Spavola: 220 μg g^{-1} ± 138; Table 1). In July when biological activity is intense, the organic P areal distribution is characterised by high concentrations on the north eastern side and in the central western section of the Fattibello lagoon, where values exceed 500 μg g^{-1}. Similar organic P concentrations can be observed in the southern part of Fattibello (Station 32) and in Spavola (Station B3), probably indicating the restricted zone of influence of the Fosse channel, characterised by a higher total P content. P concentrations are somewhat variable with increasing sediment depth. The areal distribution is modified in November with superficial peaks of organic P at the centre of the Fattibello basin, within the sub-lagoon and in the extreme eastern part of the basin (Stations 16 and 24). Concentrations are considerably lower in Spavola and tend to assume a uniform distribution.

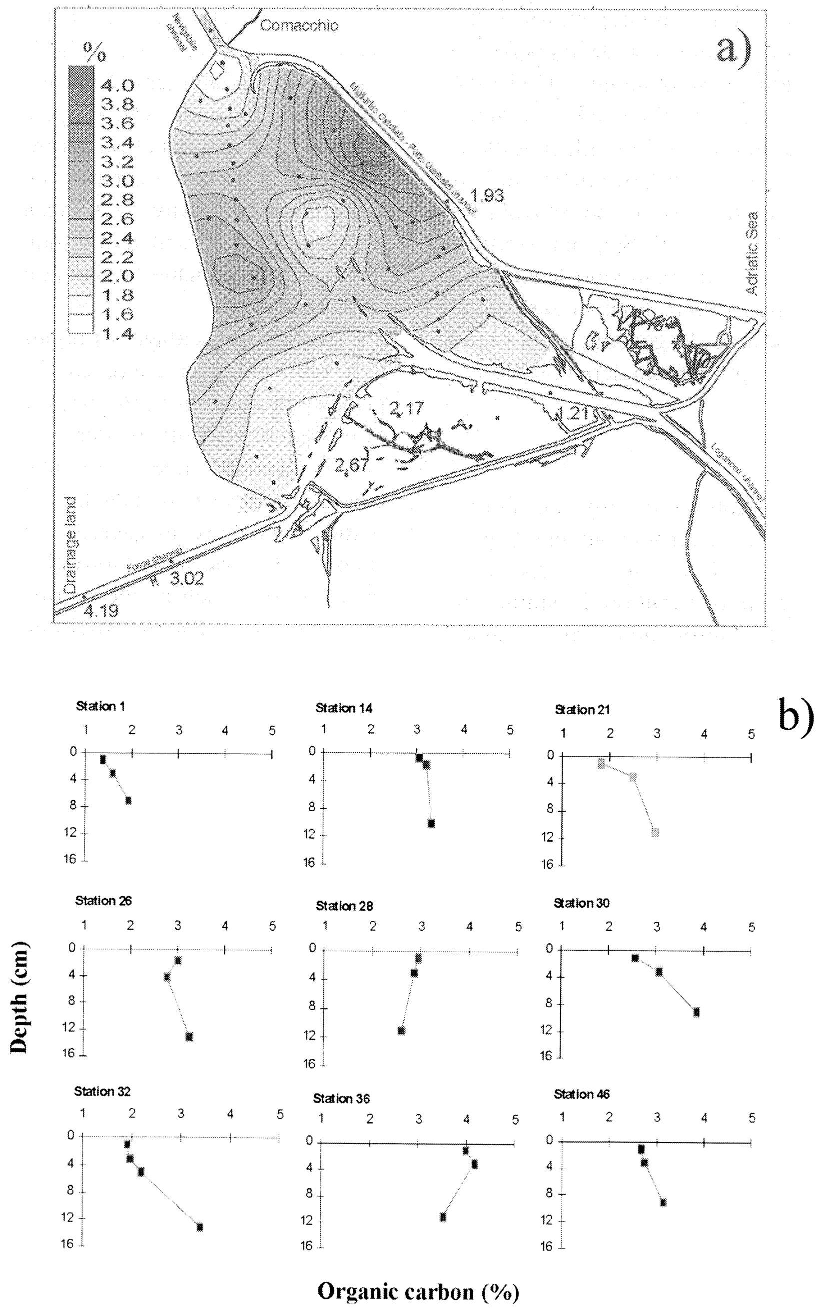

Figure 4. (a) Areal distribution of organic C concentrations in superficial sediments, July 1997. Organic C highest concentrations occur on the eastern side and in the central western area of Fattibello. High organic C concentrations and low C/N ratio values (see text) indicate the high level of primary production in the basin and the scarce input of terrigenous organic matter. (b) In July 1997, organic C concentrations of sub-superficial levels have higher mean values than the superficial ones (2.62% ± 0.72), as shown in sediment depth profiles. This indicates sudden sedimentation events with buried organic matter not yet degraded.

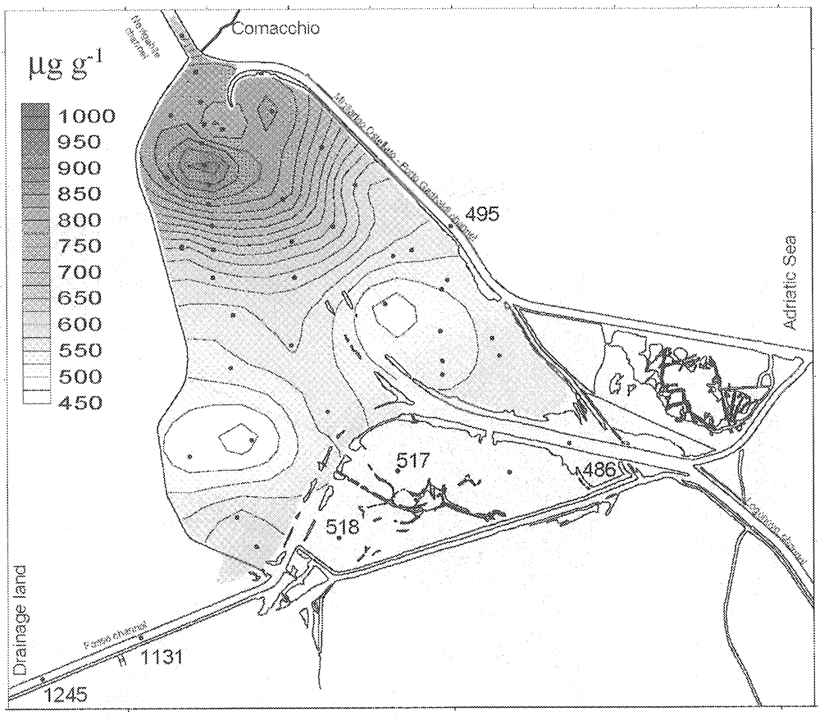

Figure 5. Areal distribution of inorganic P concentrations in superficial sediments (July 1997). The highest inorganic P concentrations occur in the northern area of Fattibello lagoon, near the Navigabile channel mouth. This highlights the importance of the nutrient loads introduced by the tributary. On the contrary, the Fosse channel, in which the inorganic P concentration in the sediments is very high, is unlikely to have an influence in this basin, as shown by lower inorganic P concentrations in the southern areas of the lagoon.

Inputs of nutrients

The high level of primary production in the basin confirms the hypothesis of a considerable inflow of nutrients from the tributaries. The basin is fed by the tributaries either directly with dissolved forms of N and P compounds or indirectly, particularly in the case of P, with forms fixed to the sediment, here predominantly inorganic. The areal distribution of inorganic phosphorus in Fattibello (Fig. 5) highlights the importance of loads introduced by the tributaries, particularly the Navigabile channel. The Fosse channel is unlikely to have an influence in this basin, except for extremely limited zones (Stations 32 and B3).

A significant input of nutrients in the basin, undoubtedly important in moments of low inflow from the land, may therefore be supplied by the system itself from bottom sediment as a result of aerobic and anoxic decomposition of organic matter. However, while the N compounds are mineralised predominantly in the inorganic soluble or gaseous form and are therefore immediately recycled in the water column, the P compounds may be partly dissolved and partly associated with the fine inorganic particulate.

Phosphorous availability

The primary production in aquatic environments is usually controlled by the availability of dissolved P compounds (Fong et al., 1993). Organic and inorganic P in the particulate fractions also play an important role. The inorganic association of P with the solid matrix may take place through adsorption on the surface of the particles or through precipitation as or with newly formed minerals (apatites, Fe and Mn oxides and hydroxides; Barbantiet al., 1994.). It is also favoured by the precipitation processes occurring during mixing of fresh and salt water (estuarine processes; Sholkovitz, 1976) at the mouths of the tributaries flowing into the Fattibello and Spavola lagoons. Both the adsorbed fraction and the fraction bound to the Fe and Mn oxides and hydroxides are considerably influenced by changes in the chemical and physical conditions. While they tend to precipitate under oxidising conditions, they tend to enter into solution under reducing conditions and be recycled into the environment.

Part of the suspended solids accumulate in the depressions of the Fattibello lagoon where, as described above, conditions of hypoxia often occur due to the

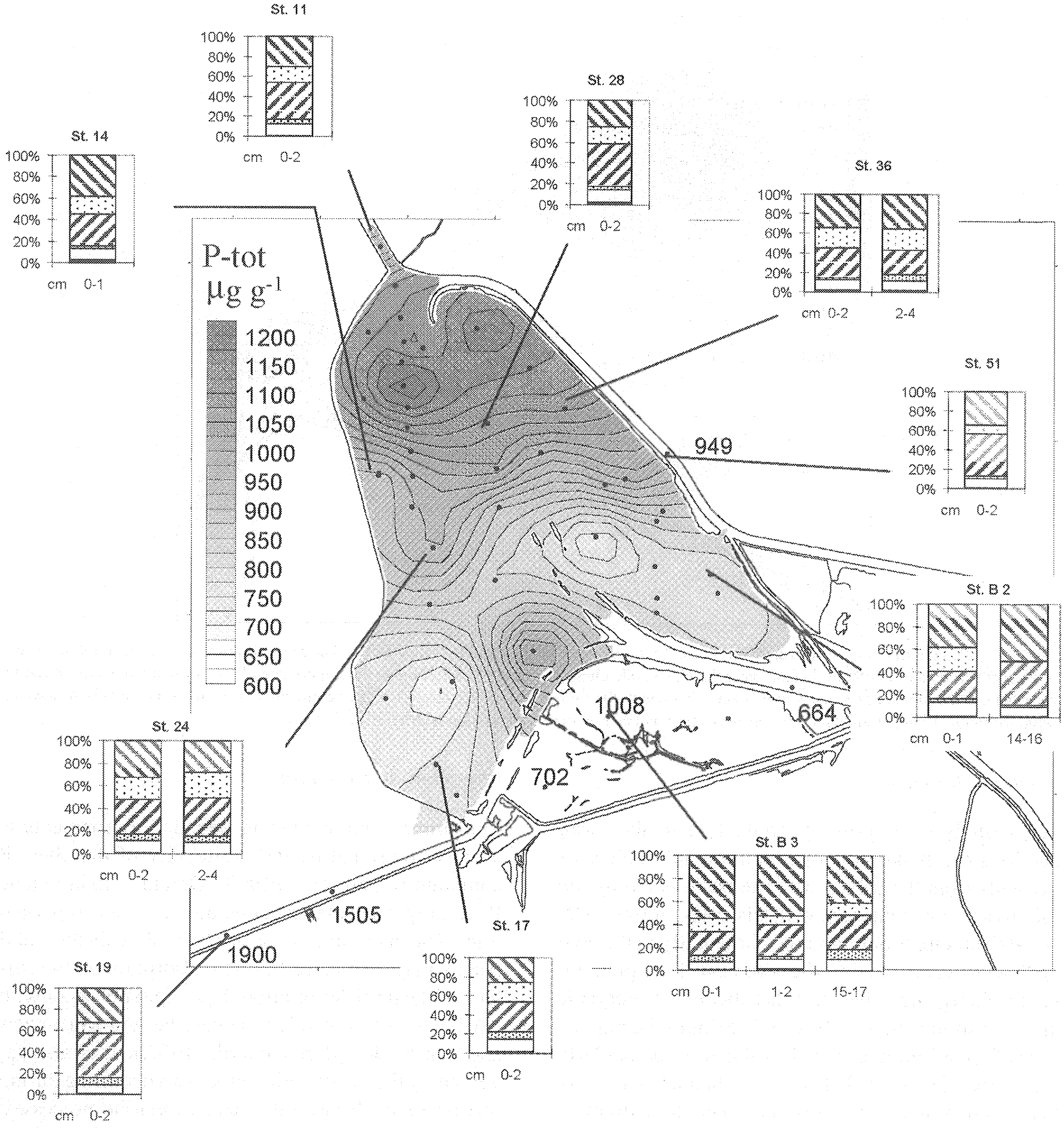

Figure 6. Areal distribution of total P concentrations in superficial sediments and histograms of P fractions in superficial and sub superficial samples in July 1997. ■ Exchangeable fraction; ☐ carbonate fraction; ☐ easily reducible fraction; ▨ moderately reducible fraction; ☐ organic and sulphide fraction; ▧ residual fraction. The mean concentration weights of easily exchangeable fractions (1.5% compared to 1.0%), of fractions bonded to the organic matter (18% compared to 13%) and of residual fractions (36% compared to 32%) are greater in the lagoon superficial samples than in the tributary channel ones. Sediment in the tributary channels is distinguished by a higher percentage of reducible fractions (45% against 34%). The higher residual fractions are in the superficial levels of Spavola lagoon.

consumption of O_2 during aerobic and anoxic decomposition of surface organic matter. Here, early diagenetic processes typical of anoxic conditions take place (Berner, 1980). These processes enable the Fe and Mn oxides and hydroxides to enter into solution, together with the substances attached to them in the interstitial water, including P (Berner, 1980; Krom & Berner, 1981; Redshaw et al., 1990). All this may be released into the water column through molecular diffusion (Eimeis et al., 2000), if the bottom waters are also anoxic. This situation can be observed in the above mentioned zones of the Fattibello lagoon as demonstrated by the fact that Mn concentrations in the surface sediment at the deepest points subject to anoxia are the lowest in the system.

Selective analyses were carried out (Barbanti, 1989; Fig. 6) to better evaluate the particulate fractionation of phosphorus in the sediments and its recycling.

The relatively marked differences between the sediment in the tributary channels and the Fattibello-Spavola lagoons demonstrate that P is easily recycled in the lagoon environment of the reducible fractions. The reducing environment in the basins evidently facilitates oxide and hydroxide decomposition and release of P into the water column for use in primary production. At sub-surface levels, the P tends to be linked to the solid matrix in more mobile forms (Fig. 6), although, as described above, the concentration in μg g^{-1} diminishes. An exception to this tendency can be observed in the south eastern part of the Fattibello lagoon (Station B2). The residual phase and the reducible phase increase at a level of 16 cm, while the phase bound to the organic matter diminishes considerably. The general tendencies observed at sediment depth confirm the hypothesis of inflows to the lagoon system capable of burying the organic matter and preserving it from more rapid aerobic degradation processes, as described above. Briefly, the fraction practically unavailable to the environment (residual fraction) does not reach 40%.

All the above demonstrates that the basin is directly and indirectly over-supplied with nutrients; that the recyclable budget is constantly increasing as a result of the retention of organic and inorganic particulate in the morphological depressions, forming a progressively larger slow release reserve able to produce ever more temporally and spatially extensive dystrophic conditions. Action to reduce this retention capacity is therefore desirable.

Statistical data analysis

A ‘Q’-Factor Analysis was applied to both the more conservative data (grain size, mineralogy, inorganic C) and to part of the less conservative data (water content, temperature, pH, Eh, organic C, total N, inorganic P, organic P, C/N ratio), in order to facilitate a concise overview of the characteristics of the superficial sediment in the two seasons, according to the trophic conditions of the basin.

Table 2. Four factor axes composition expressed as “Varimax Factor Score Matrix” (Davis, 1973). The first two factors were found to be highly significant (together satisfying 78% of the totel variability of the system) and represented the principal biogeochemical-sedimentological *‘facies’* of the whole benthic lagoon system, which can be described by combining them in various proportions. The third and fourth factors are less important, but highlight certain compositional anomalies in a number of zones with respect to the description provided by the first two. These anomalies play a fundamental role in the balance of the ecosystem

Eh	0.05	0.16	0.18	−0.03
% H_2O content	0.36	0.10	−0.17	−0.13
>0.5 mm	−0.17	0.49	0.05	0.12
0.5–0.063 mm	−0.19	0.27	0.44	−0.11
<0.063 mm	0.44	0.13	−0.16	0.18
inorganic C	−0.16	0.44	0.10	−0.01
organic C	0.27	0.16	−0.23	−0.60
N	0.26	0.20	−0.24	−0.11
C/N	0.10	0.16	0.33	0.21
inorganic P	0.10	0.07	0.10	−0.34
organic P	0.19	0.23	−0.20	−0.25
Quartz	0.25	−0.10	0.21	0.60
Plagioclase	0.17	−0.06	0.41	−0.09
Feldspar	0.13	−0.06	0.25	−0.18
Calcite	0.19	0.18	0.18	−0.11
Mg-calcite	−0.10	0.39	−0.03	−0.32
Dolomite	0.14	−0.80	0.12	0.17
Aragonite	−0.11	0.30	−0.10	0.58
Gesso	0.13	0.11	−0.07	0.33
Illite	0.23	0.07	0.25	0.16
Chlorite	0.34	0.03	19.00	0.13
Serpentine	0.13	0.05	0.13	0.01
Variance	4	33.86	11.04	2.61

Four factors were identified (Table 2), but only the first and the second ones are predominant.

Factor 1 identifies a *facies* consisting of fine inorganic particulate, predominantly silicoclastic and partly carbonoclastic, principally terrigenous and well hydrated. The organic matter (high concentrations of organic C, total N and organic P) found in this *facies* was in reducing conditions, with C/N ratios marginally higher than that of the newly formed autochthonous organic matter and therefore as yet only slightly degraded. Table 3 gives the composition of the superficial sediment of Station 14, which can be considered

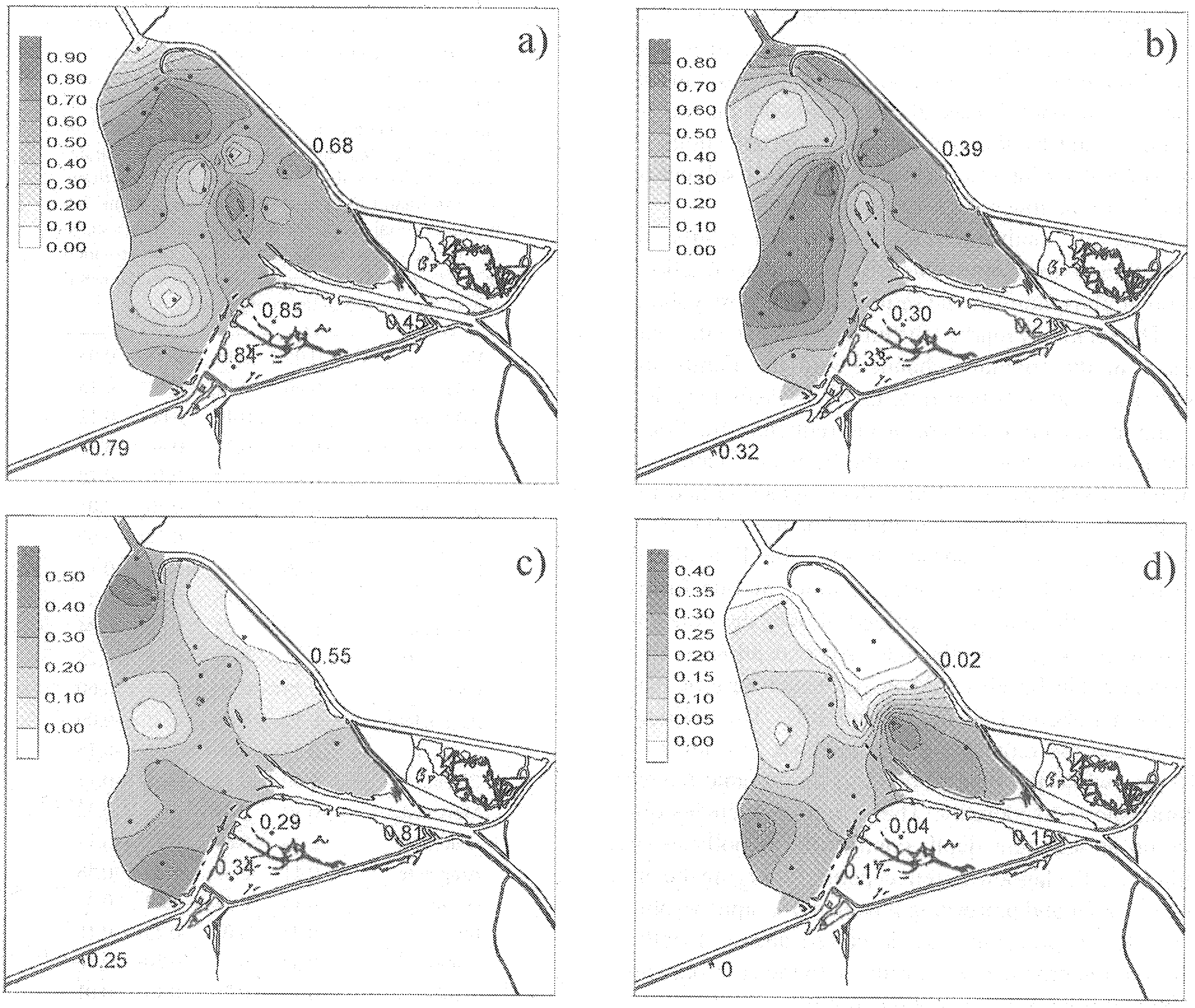

Figure 7. Areal distribution of the *facies* obtained from the Q-FA applied to superficial sediments in July 1997. (a) *facies* 1: fine silicoclastic inorganic particulate, predominantly terrigenous and well hydrated, associated with slightly degraded organic matter in reducing conditions; (b) *facies* 2: predominantly coarse biogenic fractions and finer sediments associated with not yet decomposed organic matter; (c) compositional anomaly (*facies* 3): allochthonous silicoclastic sands with degraded organic matter in reducing conditions; (d) compositional anomalies (*facies* 4): dark grey coloured area is characterised by fine fractions and aragonitic bioclasts, with lower inorganic P concentrations and high C/N values. White coloured area is characterised by high organic matter production supplied by high nutrient inputs.

an end member representative of *facies* 1. The areal distribution of this *facies* (Fig. 7a), with mud in reducing conditions and able to release nutrients over long periods, is characterised by high values in July, particularly in the morphological depressions at the base of the submerged fan at the mouth of the Navigabile channel in the Fattibello lagoon and in patches along the internal sub-lagoon channel. Significant values are also found in the tributary channels in low water periods (the Navigabile channel and Fosse channel, respectively). Values in the sediment are quite high in the Spavola lagoon, but here the bottom is more oxygenated than Fattibello and the water does not reach levels of hypoxia or anoxia. There is higher enrichment in November in the central northern depressions and the sub-lagoon channel, capable of accumulating larger quantities of fine particulate and of being at least partly transported out to sea by tidal currents.

Factor 2 identifies a *facies* consisting of predominantly coarse biogenic material (fragments of predominantly mollusc shells and constructor organisms) associated with sand or finer sediments (<63 μm) with high concentrations of Mg-calcite, aragonite and inorganic carbon. The organic matter found in this *fa-*

Table 3. Principal sample composition of each factor. St. 14 and 27 are considered end members of *facies* 1 and *facies* 2, respectively; while the others (St. 50 and St. 39) are considered samples in which the anomalies identified by factors 3 and factor 4, respectively, are better represented

July 1997		St. 14 East *Facies* 1	St. 27 Centre *Facies* 2	St. 50 Channel (*Facies* 3)	St. 39 West (*Facies* 4)
Eh	mV	−352	−344	−185	−388
pH		7.3	7.5	7.4	7.2
H_2O content	%	73.6	56	47.6	62.2
>0.5 mm	%	0.1	50.3	0.3	31.3
0.5–0.063 mm	%	0.9	11.4	34.6	9.5
<0.063 mm	%	98.9	38.3	65.2	59.2
Quartz	%	31	13	25	19
Plagioclase	%	10	4	14	6
Feldspar	%	6	0	9	0
Calcite	%	23	13	11	12
Dolomite	%	4	2	4	3
Mg-calcite	%	0	23	0	5
Aragonite	%	trace	28	0	35
Illite	%	12	11	19	9
Chlorite	%	4	2	3	2
Serpentine	%	4	2	3	2
Others	%	6	2	1	7
inorganic C	%	1.83	6.94	1.93	4.07
organic C	%	3.05	1.89	1.21	2.9
N	%	0.43	0.28	0.14	0.39
C/N		8.4	8.0	10.4	8.7
total P	$\mu g\ g^{-1}$	900	1025	664	704
inorganic P	$\mu g\ g^{-1}$	632	681	486	476
organic P	$\mu g\ g^{-1}$	268	344	178	226

cies has very low C/N values and is presumably not yet decomposed. Table 3 gives the composition of the superficial sediment in Station 27, which can be considered an end member representative of *facies* 2. The areal distribution of this *facies* (Fig. 7b) is almost specular to that of the previous *facies* and is similar in the two seasons, with the exception of the zone at the mouth of the Navigabile channel in the Fattibello lagoon where in July the values are higher. Values are low in the Spavola lagoon in both seasons.

Factor 3 identifies zones containing fractions of poorly hydrated sandy silicoclastics in less reducing conditions and with organic matter characterised by high C/N values (Fig. 7c). The sample best representing the composition of this *facies* is located in both seasons in the channel communicating with the sea (Station 50, Table 3). This anomaly is also present in significant percentages in the sediment of the submerged fan at the mouth of the Navigabile channel and less markedly in areas of the Fattibello communicating with peripheral channels (the Fosse to the south west, the Migliarino to the south east). The sandy material is apparently both land and sea derived and is deposited mainly on the bed of the channels and at the mouth of the lagoon area, site of a morphological barrier typical of all communicating zones.

Factor 4 identifies two anomalies, which were inversely correlated one to the other. The first is located in the central southern part of the basin where occurs the accumulation of poorly hydrated pelitic fractions and aragonitic bioclasts, with higher Eh values, lower inorganic P concentrations and high C/N values (Fig. 7d). Previously reworked materials with degraded autochthonous organic matter had settled in this area. The higher correlation of the southern part with factor 4 demonstrates the low impact of nutrient inputs of the Fosse channel in the lagoon. The other is located in the north eastern part of the lagoon and is characterised by primary autochthonous production supplied by nutrients transported by the Navigabile channel.

Conclusions

The biogeochemical study of the bottom in two different seasons, supported by morphological reconstruction, enabled the depositional settings and trophic conditions of the Fattibello and Spavola lagoon basins to be defined.

The first fact to emerge is the tendency for both basins – particularly the Spavola lagoon – to silt up with fine predominantly silicoclastic material deriving from the tributary channels. Energy conditions are, however, still such as to oppose this process. The shallower zones of the Fattibello lagoon are continuously oxygenated by tidal currents and wave motion, allowing numerous types of organisms to develop, including molluscs and constructors whose remains represent an important component of the sediment. The *facies* identified by factor 2 of the multivariate analysis corresponds to this type of bottom.

The considerable tidal excursion enables a salt water wedge to penetrate into the mouths of the tributaries. However, the dense water in Fattibello never completely withdraws from the sub-circular depressions, forming zones of persistent stagnant water, and favouring the localised deposition of part of the fine inorganic particulate deriving from the tributaries. These depressions are also excellent traps for the autochthonous organic matter produced in such abundance during the summer months. These accumulation mechanisms in the bottom sediment generate reserves with an extremely high content of organic matter available for recycling as nutrients in the basin through early diagenesy processes and benthic flux through molecular diffusion. The stagnant water in these depressions rapidly becomes hypoxic or anoxic, favouring further release, particularly of phosphorus and possible contaminants. The general anoxia in sediments may be widely transmitted to the sediment–water interface during algal blooms and consequently to the bottom water, especially in Fattibello. The most typical expression of the *facies* identified by factor 1 is represented by this type of bottom, largely determined by morphological problems, which can be resolved by removing the obstacles preventing the salt water wedge from withdrawing completely during low tides.

This was taken into account during local authority channel dredging when an attempt was made to isolate the Spavola lagoon from direct inflow from the land, thus preventing silting up of the basin. In addition, a number of dredging operations were carried out in Fattibello with immediate benefits to the system in terms of oxygenation, including during the summer months.

One serious problem which can only be resolved in the long term concerns the need to reduce the inflow of nutrients from the land, clearly demonstrated by the extremely high micro and macro algae production in the basins (in the case of Fattibello, the loads in the Navigabile channel must be reduced). Even if the eutrophic effect on the lagoons is reduced by improving circulation, the only real probable effect is to diminish the filter action exerted by these basins in relation to the sea, thus increasing the anthropogenic pressure on the already sorely taxed coastal zones of the Po.

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Changes in point and non-point sources phosphorus loads in the Thau catchment over 25 years (Mediterranean Sea – France)

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Key words: phosphorus loads, point source, non-point source, land-use, erosion, GIS, Thau lagoon catchment, Mediterranean coast

Abstract

In Thau coastal lagoon, phosphate concentrations have decreased by 89% from 1971 to 1994. The present relatively long term (over 25 years) study compares changes in the contribution of point (PS) and non-point sources (NPS) total phosphorus (P) loads. The analysis of the distribution of the sources in comparison with the changes in the phosphate concentrations in the Thau lagoon aims to point out their relative impact in order to create a sustainable management plan for this system. This is needed, firstly because water quality supports shellfish farming, which is the main economical activity of the basin. Secondly, because the population is planned to increase by 40% between 1995 and 2020 thus leading to an increase of urban pressures. PS P loads, represented by discharges by wastewater treatment plants, have increased by 143% while NPS P loads, both represented by export from lands and loads from non-connected population, have decreased by 64%. Despite important changes in land-use by an exceptional decrease of vineyards areas (−12.5%), domestic effluents main contribute (>60%) to both PS and NPS P loads and seem to be more implicated in the decrease of phosphate concentrations in the Thau lagoon, probably because of the different phosphorus forms engaged.

Introduction

Since the industrial revolution, the N and P contamination of natural systems has been highly correlated with human activities (Vollenweider, 1971). Both industrial, agricultural and sewage from population contribute to the enrichment of natural waters. Furthermore, because of their poor water turnover, Mediterranean coastal lagoons are particularly responsive to fluxes from catchments (Frisoni, 1987). For example, during the 1970s, severe anoxia events and eutrophication occured in French Mediterranean coastal lagoons (Vaulot & Frisoni, 1986). In particular, in 1987 in the Thau lagoon, shellfish farms suffered from considerable losses estimated at 5 million Euro because of these events (Garrabé & Cabassut, 1989). Despite this and the lack of nutrients limiting politics, a 89% decrease of phosphate concentrations occured in the water column of the Thau coastal lagoon between 1971 and 1994 (La Jeunesse, 2001).

This observed trend on phosphate concentrations can be explained both by a new ecological equilibrium in the Thau lagoon and by changes in total phosphorus (P) quantities generated by human activities on the Thau catchment. The study considers the second hypothesis by analysing the changes in point (PS) and non-point source (NPS) P loads produced. PS-P loads studied are represented by loads generated by wastewater treatment plants. NPS-P loads studied are represented by export from lands (both cultivated,

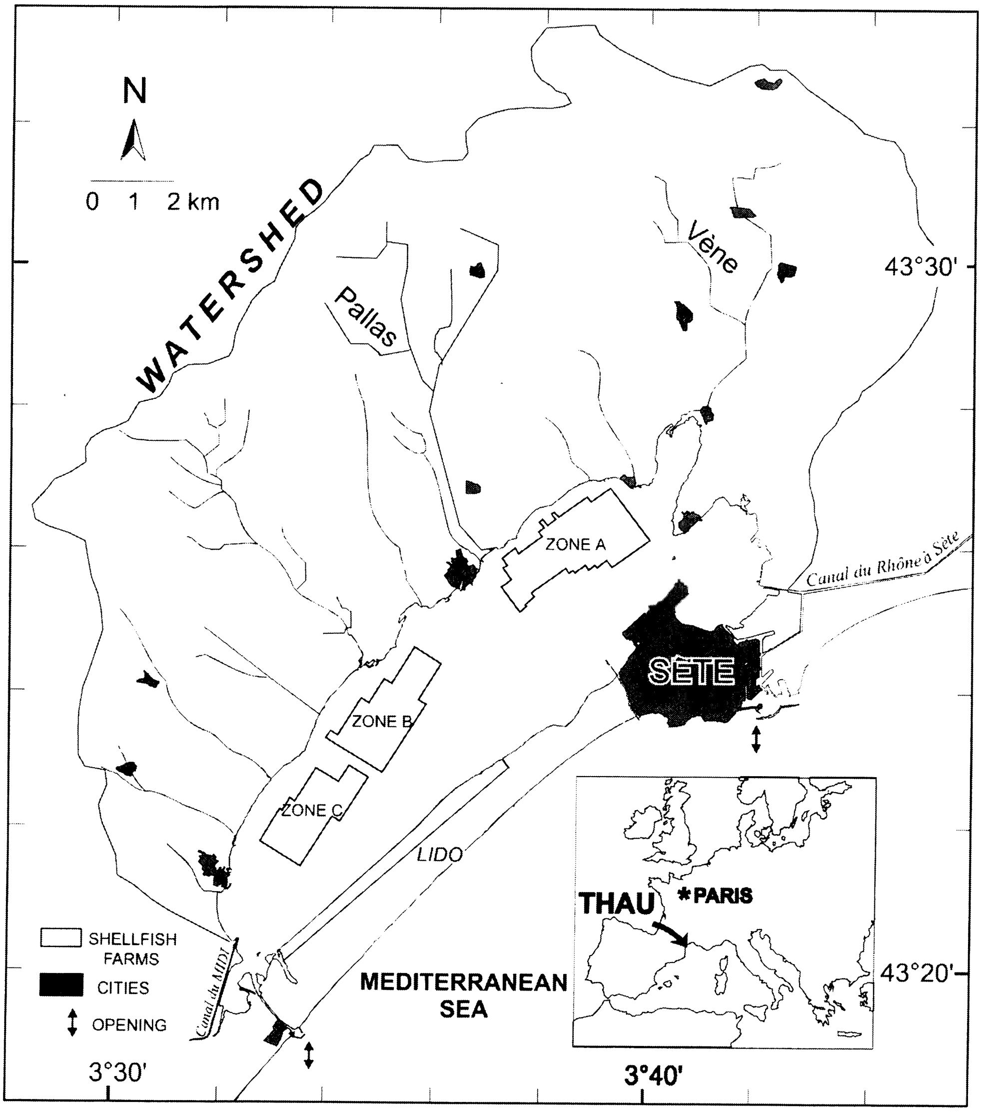

Figure 1. The Thau coastal lagoon and catchment.

non-cultivated and urbanised) and also by loads from the non-connected population. A long-term study is necessary to evaluate the impact of PS and NPS-P loads on the Thau lagoon in order to support decisions for a sustainable management of the system. The latter is required, firstly because water quality has to continue to support shellfish growth and secondly because from the French National Institute of Statistics and Economical Studies a 40% population growth is planned between 1995 and 2020 in the Thau region. In fact, this paper focuses on NPS-P loads and it complements an analysis of PS P loads presented elsewhere (La Jeunesse, 2001). And, even if nitrogen is the first limiting factor of primary production in the Thau coastal lagoon (Vaulot et Frisoni, 1986; Mazouni et al., 1996), the study of nitrogen trend has not already been done. P loads produced by sewage depend on population and on the proportion of effluents treated by wastewater treatment plants. As sewage are discharged directly into rivers in the Thau catchment and because of the Mediterranean weather characteristics impact already described, it is assumed that all wastes should reach the lagoon within a year.

The impact of catchment on P transfer in surface waters depends on regional geology, erodability and land-use (Dillon & Kirchner, 1975), catchment size

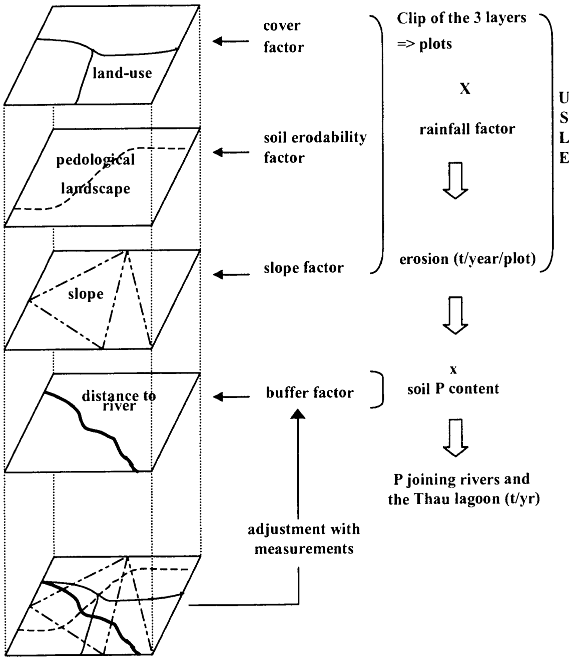

Figure 2. GIS-based P export model principles.

(Bolton & Ward, 1993), shape and topography (Osborne & Wiley, 1988), precipitations (Wichmeier & Smith, 1978) and soil P content (Pote et al., 1996). Interpretation of P transport factors is usually simplified assuming erosion controls particulate P movement and runoff dissolved P movement (Sharpley et al., 1993). Firstly, the Thau catchment is controlled by Mediterranean weather conditions characterized by hot and dry summers and by intense rainfall events where autumn storms can represent up to 75% of the annual rainfalls (Tournoud et al., 1997). Secondly, analysis of P forms in the Thau catchment's soils following the Olsen method (Olsen et al., 1954) show particulate forms represent more than 96% of the P soil content. Hence, not only particulate P is the major proportion (75–90%) of P transported from cultivated land (Sharpley et al., 1987) but also it is mainly exported during the largest storm events (Johson et al., 1976). Thus, it is assumed here that NPS-P loads on the Thau catchment are mainly produced by soils' rainfall-induced erosion.

As required data for export coefficients approaches (Norvell et al., 1979; Reckhow & Simpson, 1980) were not available on the Thau catchment within the study period, a modelisation approach is used. As deterministic models of erosion require too many data usually not available at the catchment scale (Benkhadra, 1997), an empirical erosion model is used: the Universal Soil Loss Equation established by Wischmeier & Smith (1978). This model is linked here to a GIS as it provides (1) large amounts of input data required by the model and reduces uncertainty caused by spatial averaging (Lenzi & Luzio, 1997), (2) identification of environmental sensitive areas and (3) appraising alternative management strategies and programs (Tim et al., 1992).

Because of everything that is said, the objective of the present study is to discuss the contribution of the changes of human activities during the past 25 years on the Thau lagoon's phosphate concentrations. An analysis of land-use change on erosion and phosphorus export will be done. Then, an estimation of the impact of population growth and sewage improvement will be discussed to compare PS and NPS-P load changes and its eventual implication in changes in the Thau coastal lagoon's phosphate concentrations.

Description of the study site

The catchment covers 250 km^2 including a total population of 80 000 inhabitants with half of them concentrated in the city of Sète (Fig. 1). The total population doubles each summer through tourism (SIEE, 1992). As 40% of the catchment is occupied by vineyards, wine and raisin production is the main agricultural activity. Large areas of soil surface remain uncovered, even during the growing season, due to the wide spacing of the plants and weedings. There is only a small industrial activity concentrated in the city of Sète mainly represented by two units of fertilizers production.

The Olsen fraction (bioavailable forms, Oslen et al., 1954) represents in the soil of the Tau catchment 3.10% (±S.D. 1.19%) of P forms and dissolved inorganic phosphorus represents 0.18% (S.D. 0.09%). Therefore, >96% of P element is in particular form. Then, as already observed in other sites (Svendsen et al., 1995), there is no significant relation between P soil content and land-use (ANOVA, p=0.985) or pedological landscape definitions (p=0.3236). Thus, a mean soil P content of 745 ppm (±S.D. 483 ppm) measured on 33 samples collected in July 1998 is used.

The Thau coastal lagoon covers 75 km^2 and it has a mean depth of 4 m with a maximum of 11 m. Its volume approximates 300 Mm^3 and the tide does not exceed 30 cm. It receives freshwater from many small and mostly non-permanent streams from the catch-

Table 1. P loads generated by the population

P loads from sewage	1971	1996
Connected population to a wastewater treatment plant	13%	92%
P loads from connected population (t/yr)	12.4	30.1
P loads from non-connected population (t/yr)	77.0	11.9

ment. Shellfish-farming covers one fifth of the total area with an annual production of shellfish of approximately 15 000 tons (Deslous-Paoli et al., 1998). In the Thau coastal lagoon, shellfish-farming and fishing are the designated priorities because of both exceptional nutritional potentialities of the lagoon and their major economic contribution to the region.

Methods

Annual P loads generated by each inhabitant, resident or tourist, are assessed by multiplying the daily P waste (function of detergent consumption) by 365 days. For the connected population, assessed with water consumers bills, the treatment rate of wastewater treatment plants, differing with process used (activated sludge, natural lagoons...) and chronic dysfunctioning occurring, is implemented to loads generated. P loads from non-connected population are supposed to have no treatment and to contribute entirely to diffuse pollution (La Jeunesse, 2001).

The Universal Soil Loss Equation (USLE, Wischmeier & Smith, 1978) has been implemented to plots resulted from clips between three layers: land-use, pedological landscape and slope (Fig. 2). This permits us to use not average plot values for each factor (respectively C, K and S) of the USLE. The USLE enables us to calculate quantities of sediments exported by erosion from each plot. All the P contained in these sediments is assumed here to be exported. A buffer effect during transfer is assumed here to be proportional to the distance to the river, concept already applied to P transfer on a more humid zone (Lake Geneva sub-catchment: Bouchardy, 1992). Four classes are arbitrarly defined: 0–100 m, 100–200 m, 200–500 m and >500 m to which corresponds, respectively, to 80%, 50%, 20% and 0% of P joining the stream.

The rainfall data used are from June 1995 to May 1996, chosen as a mean of average rainfall (a mean of a wet and a dry year). The rainfall intensity factor used resulted in a mean of the factor calculated with the region's rainfalls over 20 years (Pihan, 1979).

ArcInfo and ArcView 3.1 GIS software have been used to manipulate the data in digital form and determine and analyse USLE parameters directly from maps. A Digital Elevation Model has been extracted from the 1:50 000 National Geographical Database, BD-Carto (IGN, National Geographic Institute, 1994), to obtain slopes and superficial hydrosystem covers. Pedological landscape layer was extracted from the 1:250 000 Regional (Languedoc-Roussillon) Database (Robbez-Masson et al., 2000). The land-use is the interpretation of ortho-rectified aerial photographs (IGN) of summer 1971 (1:17 000 black and white paper photographs) and summer 1996 (1:25 000 colour CD-Rom and paper photographs). Ground validations were carried out in 1998 for land-use plots which did not changed between 1996 and 1998. Then, National and Departmental Agricultural Statistics (Recensement Général Agricole: 1970, 1979, 1988; Direction Départementale de l'Agriculture: 1996) indicate the main cultivated areas at a district scale in 1970, 1979 and 1988.

Urban areas are also concerned by runoff due to some wastes and detergents used to clean the cities. P quantities exported are assessed to approximated 2.5 kg P/ha of urban areas/year (Vollenweider, 1971).

Results

The connecting population to a wastewater treatment plant increased from 13% to 92% between 1971 and 1996 (Table 1). Because of this in particular, PS P loads have doubled and NPS P loads have decreased by 85%.

The main evolution of land-use in the Thau catchment is the important decrease of vineyards which represented 48.4% of the catchment (Table 2) in 1971 against 35.6% in 1996. Those 12.5% areas lost have been built upon 4.8% or abandoned (fallow-land and forest-garrigue: 4.6%) and or replaced by cropland

(3.2%). National statistics (Recensement Général Agricole: 1970, 1979, 1988) indicate the decrease of vineyards occurred linearly during the study period (r^2=0.994).

The increase of urbanised areas induced a proportional increase of urban run-off reaching now 6.9 t/yr and representing 10% of the P loads assessed here (Table 5).

The GIS-based erosion model run for 1971 and 1996 estimates an average decrease of 27% in sediments exported from plots by erosion leading to a 21% decrease in P export because of land-use changes (Table 3). The S.D. attached to P export is here due to the S.D. on measurements on soil P content.

The spatial analysis of the contribution of land-uses show the importance of vineyards (Table 2) representing more than 80% of P export both in 1971 and 1996. The decrease of vineyards lead to a decrease in its contribution by 5% replaced by an increase in cropland (mainly wheat) contribution. The spatial analysis of the contribution of pedological landscape importance on P export pointed out 45.1% of vineyards areas on the catchment represent more than 73% of P export (Table 4). This has not changed significantly within the study period. The forest-garrigue cover, which has not significantly changed within the study period either, exports very low P quantities (2–4%) in proportion to its surfaces (>9000 ha). Hence, the vegetation density of forest-garrigue cover represents a high protection against rainfall erosion.

The changes for each source of P load studied and its changes between 1971 and 1996 are presented in Table 5.

Discussion

The assessment of loads coming from the non-connected population are maximum because of the assumption its effluents entirely join superficial hydrosystem. But, in 1996, even if there are no official registration of existing septic tanks and their treatment efficiency, because of the legislation (Directive no. 91/271/), we can assume non-connected houses have their own septic tanks that work accurately (that is to say without any overflowing in the superficial hydrosystem, Philippi et al., 1992). This hypothesis led to an overestimation of NPS-P loads. Without these loads from the non-connected population, the sum of P loads generated by urbanization (the connected population and urban runoff) would be 37 t/yr. Measurements at outlets on the Thau catchment approximate 48 t/yr of total phosphorus (S.D.: 50%, Tournoud et al., 1997). Thus, PS P loads should represent 11 t/yr, that is less than results (18.1 t/yr) of the model developed here. This latest probably overestimates erosion and P export. This can be due, firstly, to an overestimation of erosion with the implementation of the USLE at the plot scale or, secondly, to an underestimation of the buffer coefficients at the catchment scale. The USLE has been established by Wischmeier & Smith (1978) on the basement of available data on 10 000 land-use plots situated in Northern America. This prompts to criticize the universal use of this equation. To discuss the implementation on the Thau catchment, it is possible to compare its results with the available measurements in the Mediterranean agronomical experimentation pilote site situated 30 km close to the Thau catchment (Roujan catchment). Two plots having slopes situated between 5% and 15% have been monitored. Average quantities of sediments eroded during a dry year (1995) are 8.4 t/yr and 30.6 t/yr during a wet year (1997; Andrieux et al., 1998). The implementation of the USLE in 1996 (a humid year), for the same range of slopes, evaluates the average loss to 23 t/yr. Thus, the overestimation does not occur at the plot implementation scale for this range of slopes. But, considering the sensitivity of the USLE to intensive rainfalls and steep slopes (Whischmeier & Smith, 1976), extremes conditions could also generate over-estimations. Then, measurements at the outlet of one Thau sub-catchment (Pallas river, Fig. 1) in 1995 and 1996 shows an export of $3.4.10^3$ t/yr of suspended particulate matter (SPM, Tournoud et al., 1997). This represents an average SPM export of 0.7 t/ha/yr. This is approximately 10 times less than results of the implementation of the model proposed here (Table 3). This means clearly the model could be improved by analysis of buffer capacity of the Thau catchment, in cultivated and natural areas as in rivers beds.

Between 1971 and 1996, the very dynamic changes of land-use do finally not lead to a spectacular change in the export of phosphorus in the Thau catchment. If the model answered to the assumption of the implication of these changes in the general decrease of P loads in the catchment, stronger impacts on erosion could have been expected. In fact, the implementation assumes all the variables remain constant except land-use. These assumptions probably obscured some changes in important variables as fertilization and agricultural techniques. But, on this semi-long term study period, it seems difficult to approach fertilization

Table 2. Evolution of land-use and total phosphorus losses

Cover	1971			1996		
	Surfaces (ha)	% Watershed	% of TP Exported	Surfaces (ha)	% Watershed	% of TP Exported
Vineyards	13 471	48.4	**91.6**	9983	35.9	**83.6**
Forest-Scrub	9094	32.7	**2.4**	9246	33.3	**3.5**
Fallow-Land	1580	5.7	**0.3**	2696	9.7	**0.7**
Urban areas	1213	4.4	–	2548	9.2	–
Cropland	860	3.1	**2.3**	1756	6.3	**8.1**
No Cover	495	1.8	**2.3**	399	2.0	**2.9**
Orchards	179	0.6	**0.9**	208	1.4	**1.0**
Quarries	70	0.3	**0.2**	65	0.2	**0.2**
Marshy Land	834	3.0	–	736	2.0	–

Table 3. Results of the GIS-based total phosphorus export model

	A (t/ha/yr)		TP (kg/ha/yr)		TP (t/yr)	
	Average	S.D.	Average	S.D.	Sum	S.D.
1971	11.5	37.2	1.9	7.1	23.0	14.9
1996	8.4	20.2	1.5	5.6	18.1	11.7

as far as nutrients stocks in soils have been constituted because of fertilization at least since the 1950s with demonstrated impacts until 60 cm deep (Comlan, 1996); which thus could not significantly decrease without any strong decreases in fertilization during many years. Then, agricultural practices are very difficult to generalize at the catchment scale and could be tackled only with many field studies. Despite all these things, on one hand, the present study pointed out the necessity of a fine identification of land-use and, on a second hand, it firstly indicated some clear 'critical source areas' (Gburek et al., 1996) within the Thau catchment. Between 1971 and 1996, changes in the two main human activities of the Thau catchment, wine production and urbanization, have provided a 42% decrease of P loads production. Despite exceptional loss of vineyards, the main quantities lost are because of the improvement of sewage collection and treatment. But, the most important change between 1971 and 1996 is probably the new distribution of PS and NPS. NPS were representing 90% of loads entering the lagoon in 1971 and 55% in 1996, with 67% due to non-connected population in 1971 and 18% in 1996, or probably less, as explained above. Furthermore, erosion from lands contribute only by 27% to P loads entering the lagoon. In addition, P quantities (mainly particulate) generated by erosion are only half as bioavailable than those from sewage which is mainly composed of dissolved inorganic phosphorus (Dorioz et al., 1997, 1998). Because of all is said, P loads from erosion of agricultural and natural lands are less implicated is the availability of phosphorus quantities in the Thau lagoon than loads from sewage. Then, it is necessary to consider inputs to the lagoon especially with regard to urbanization pressures given the population growth predicted by 2020 in the Thau catchment as in coastal zones in general (La Jeunesse et al., 1999).

In the Thau coastal lagoon–catchment system, phosphate concentrations have decreased by 89% between 1971 and 1994, thus much more than the decrease of P loads generated on the catchment. Either the assessement of P loads produced by the catchment has underestimated the decrease or some other variables are controlling changes in P availability in the lagoon. In fact, shellfish farms stocks have more than doubled between 1970 and 1995 (Deslous-Paoli et al., 1998), with an important increase of requirements leading to a decrease of the Thau lagoon eutrophisation (Deslous-Paoli et al., 1998). Then, even if nowadays because of legislation the two fertilizers units of the Sète city do not discharge any waste in

Table 4. Phosphorus critical source areas analysis on vineyards areas

Pedological landscale type	Silt (%)	K factor	% of the catchment	% of vineyards	1971 contribution of P export (%)	% of vineyards	1996 contribution to P export (%)
1	59.0	0.44	**5.6**	8.3	**15.8**	9.0	**22.3**
2	44.6	0.31	**12.2**	22.0	**11.6**	23.9	**19.0**
3	29.4	0.12	**10.2**	14.2	**16.3**	11.6	**10.1**
4	29.3	0.12	**17.1**	22.9	**31.1**	22.8	**21.7**
		TOTAL	**45.1**		**74.8**		**73.1**

The K factor depends on the composition of the upper soil's layer (sand, clay, silt and organic matter) but is much more linked to the proportion of silt (Wischmeier & Smith, 1978). For more information about the 24 pedological landscape types describing the Thau catchment upper soil's layer, see Robbez–Masson et al. (2000).

Table 5. Contribution of point (PS) and non-point sources (NPS) to TP (P) loads

Total Phosphorus (t)	Type of pollution	1971	% of total	1996	% of total	Changes (%) 1971–1996
Connected population	**PS**	**12.4**	**10**	**30.1**	**45**	143
Non-connected population	NPS	77.0	67	11.9	18	−85
Cultivated and natural lands	NPS	23.0	20	18.1	27	−21
Urbanised areas	NPS	3.3	3	6.9	10	109
TOTAL	**NPS**	**103.3**	**90**	**36.9**	**55**	−64
	TOTAL	**115.7**		**67.0**		**−42**
Changes in the Thau lagoon phosphate concentrations (1971–1994)						**−89**

the Thau coastal lagoon, as the legislation date back to the 1970s, the possible changes during this period should be studied.

Conclusion

The changes in both PS and NPS-P loads provided a 42% decrease of quantities entering the lagoon. The loss of vineyards areas on the Thau catchment, a high erosion risk culture, contributed to the general decrease. Despite these changes in land-use, the decrease of P loads is mainly due to the improvement of collection and treatment of sewage. Moreover, the impact of reducing NPS-P loads generated by erosion could be delayed because of a different bioavailability between P forms export by soil erosion and those from sewage (i.e. particulate and dissolved forms). Therefore, long term studies on catchment are valuable in indicating the ranking of impact by different nutrient sources on water quality. This confirms the importance of the accuracy of land-use cover for which data from aerial photographs represent a good approach. And, the GIS tool used for the study is now available to provide a management support as it pointed out criticize sources areas because of urbanization and pedological landscape characteristics. However, to produce more accurate estimates of quantities exported and also in order to have a better predictive capability, the model developed here requires buffer analysis of both sediments and P transfer as a study of changes in fertilization. Then, a 40% population growth is planned between 1995 and 2020 in the Thau region. This will induce an increase in urban pressures and surely a decrease of arable and natural lands. Thus, considering the distribution of PS and NPS-P loads, it is necessary to manage population growth and sewage without affecting shellfish farm requirements and causing eutrophication problems as those occurring in the 1970s. The GIS based model developed here is valuable for improving and accomplishing the Thau Integrated Coastal Zone Management cooperative initiatives as far as it has described critical source areas. It is considered that the main objective should be to maintain in the lagoon water phosphate concentrations at a level similar to the present one which finally shows a good equilibrium between the shellfish farms' requirements and preventing anoxia events.

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E. Orive, M. Elliott & V.N. de Jonge (eds), Nutrients and Eutrophication in Estuaries and Coastal Waters.

Nutrients related to the hydrologic regime in the coastal lagoons of Viladecans (NE Spain)

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Key words: coastal lagoons, eutrophication, phosphorus loading, water quality management

Abstract

This study presents the major water characteristics of the Viladecans coastal lagoons in the Llobregat Delta (NE Spain), and the interpretation of their functional relationships with freshwater discharges. Ranges of conductivity show that Murtra, Remolar and Vidala are strongly influenced by freshwater. A second group (Reguerons, Braç Vidala) is characterised by brackish water with intermediate values of conductivity. In contrast, the lower part of the water column of Remolar and Vidala is quite salty water. A permanent pycnocline is established causing meromixis. Finally in Sant Climent a great variation is observed from fresh to seawater. Nutrient inputs are related to freshwater discharges. The range of DIN/SRP ratios observed (0.1–71) shows that a great variety of biogeochemical processes take place in these lagoons. High DIN/SRP ratios occur in water masses receiving high freshwater discharges (Murtra, Remolar and Vidala surface). In contrast, low DIN/SRP ratios correspond to water masses (Reguerons, Braç Vidala, Sant Climent) receiving water discharges from the groundwater and to those in the lower part of the water column of Remolar and Vidala. The SRP and DIN concentrations of the water in Remolar, Vidala and Murtra are very high (higher than any other with similar morphological characteristics in Spain). However, the trophic state of Reguerons, Sant Climent and Braç Vidala is lower. The data presented indicate that the Viladecans coastal lagoons are in a hypereutrophic state, as the result of receiving over a long period of time nutrient discharges associated to man-made freshwater inputs higher than their carrying capacity or capacity to metabolise the nitrogen and phosphorus discharges. Remedial measures should include reducing nutrient discharges into the lagoons and favouring water turnover in the lagoons.

Introduction

Many estuaries are degraded all around the world because of industrial, agricultural, and urbanisation activities (Pearce & Crivelli, 1994; Mesnage & Picot, 1995; Nixon, 1995). In highly developed countries, all these human activities can affect the characteristics of the water in their estuaries. One of their most frequent impacts is eutrophication due to increased nutrient discharge and water flow regulations (McComb & Humphries, 1992; Sfriso et al., 1992; Valiela et al., 1992). Changes in both the absolute and relative concentrations of nitrogen and phosphorus are good indicators of the intensity of the eutrophication process and of disturbances in the cycling of these elements (Nixon et al., 1986; Thybo-Christensen et al., 1993). Changes in the water flows can also be responsible for estuarine degradation (Lavery, 1997). In many cases, eutrophication is originated because of both nutrient discharges associated to changes of the water flows.

Estuarine and coastal eutrophication is a progressive process, which causes hypoxia and anoxia in many estuarine areas. The intensity and persistence of the oxygen depletion is directly related to the nutrient discharges and inversely to the water turnover of the

Table 1. Morphometric characteristics of Viladecans coastal lagoons

Coastal lagoons	Volume (m^3)	Maximum length (m)	Maximum width (m)	Maximum depth (m)
Sant Climent	92 500	1500	75	1.7
Braç Vidala	122 500	1400	35–70	3.2
Remolar	105 440	930	20–100	4.8
Vidala	26 500	300	45	3.5
Reguerons	800	70	20	0.8
Murtra	15 300	600	9–30	1.4

water of the estuaries (Rabalais et al., 1985). Vertical gradients of salinity contribute to create anoxic zones in the deeper part of the water column in coastal lagoons perpendicular to the coastline and to accumulation of organic matter in the sediments (Lopez et al., 1984).

The coastal lagoons in the Delta of the River Llobregat (Viladecans, in the southern limit of Barcelona city, NE Spain) constitute a paradigmatic case because they received sewage discharges from non-point sources due to agricultural activities, and point sources due to industrial and urban uses for many years. Now, the coastal lagoons are still strongly influenced by these discharges and limited in their water exchanges with the sea due to silting and stabilisation of sandbanks in the mouths of the lagoons. This paper presents a description and a classification of the major water characteristics of the lagoons in the Llobregat Delta and the interpretation of their functional relationships with freshwater discharges. It also suggests management proposals to improve the water quality of the lagoons.

Materials and methods

Viladecans is located south of Barcelona city, in the alluvial plain formed by the delta of the Llobregat river. This area includes a number of coastal and inland lagoons, which were part of old river mouths and wetlands of the delta plain. In this area, six lagoons were sampled: Reguerons, Sant Climent, Murtra, Remolar, Braç Vidala and Vidala (Fig. 1). Braç Vidala is separated from Remolar by a compacted soil dike. New, excavated basins were created artificially for drainage (Sant Climent). Most of the lagoon areas are shallow (less than 2 m) but in some small localities a deep-water column is established (between 2 and 5 m) (Vidala, Braç Vidala, Remolar). Table 1 summarises the major morphometric characteristics of the water masses studied (Hurtado & Lucena, 1997). Six water masses (surface and deep water) were sampled seasonally during 1996–1997 and twice during 1998. Temperature, pH, conductivity (referred to 25 °C) and dissolved oxygen were measured *in situ* during the day hours. Vertical profiles of these parameters were recorded every 50 cm in the water column with a multiparametric recorder. Water samples were collected from a boat with a water-sampling bottle and were kept at 4 °C for analysis during the next 3 days. Standard methods (APHA, 1989) were used for analysis of different forms of dissolved inorganic nitrogen, soluble reactive phosphorus (SRP), suspended matter, major dissolved anions and cations, and phytoplankton chlorophyll *a*.

Results

Mineral content

The mineral content (expressed as conductivity) observed in the lagoons ranges between 3 and 48 mS cm^{-1} (Fig. 2). In the deep part of the water column of Remolar and Vidala the conductivity is relatively high and shows small fluctuations (34–48 mS cm^{-1}). In contrast, the conductivity in Reguerons, Murtra, Braç Vidala, Sant Climent and the upper part of the water column in Remolar and Vidala lagoons is relatively low (less than 20 mS cm^{-1}), with the sole exception of Sant Climent near the sea where it can reach high values (over 40 mS cm^{-1}) during some periods of the year.

The temporal variations of conductivity are relatively low in Murtra, Remolar, Vidala and Braç Vidala, while they are higher in Reguerons and Sant Climent (Fig. 3). The difference between these two groups of lagoons is that in the former group a quite constant influence of salt and freshwater is maintained while in the second group an outstanding seasonal variation of some processes dominates the water balance. In the case of Reguerons, evaporation takes the leads compared to water inputs and conductivity increases during summer months in this shallow lagoon. In Sant Climent, conductivity increases are related to marine water inflows during sea storms in winter.

Based on the concentrations of the major ions, the lagoons Reguerons and Braç Vidala are dominated by

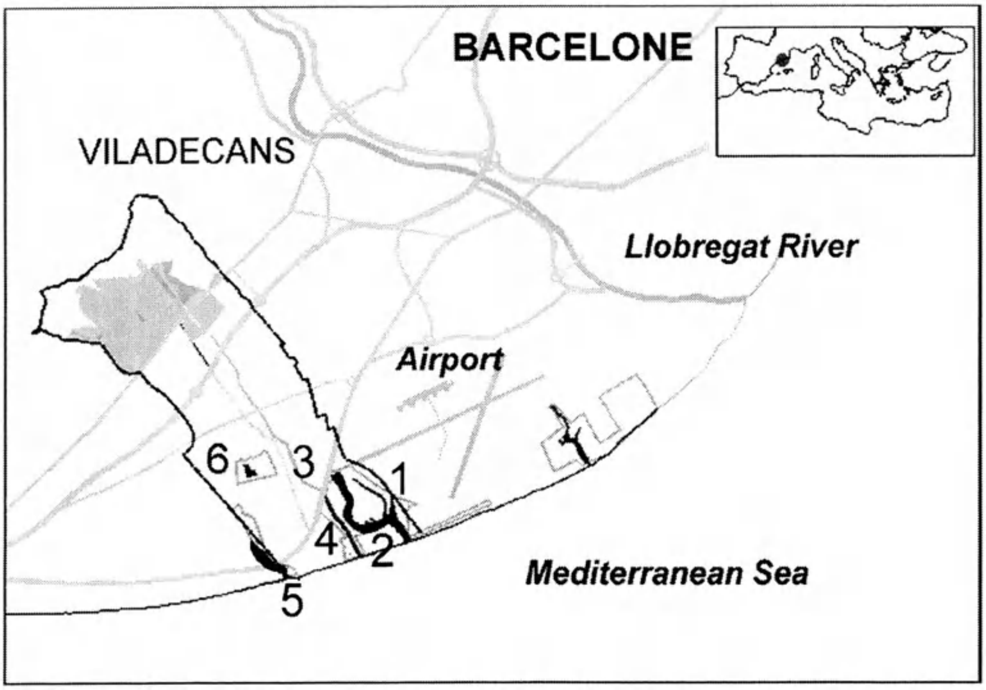

Figure 1. Map of the Llobregat River Delta showing the location of the coastal lagoons of Viladecans, and the sampling sites. 1: Remolar; 2: Vidala; 3: Braç Vidala; 4: Sant Climent; 5: Murtra; 6: Reguerons.

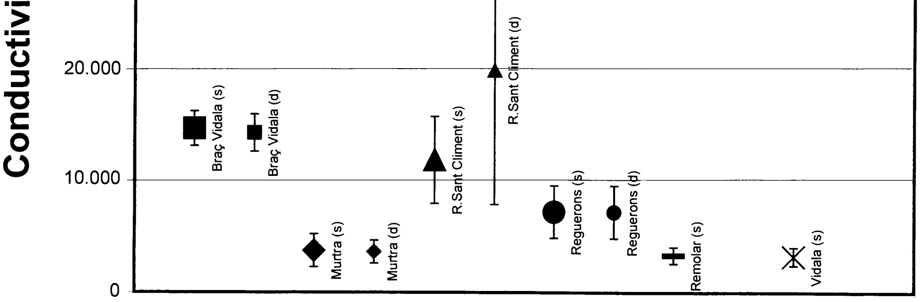

Figure 2. Ranges of conductivity measured (μs cm^{-1}) in the coastal lagoons of Viladecans (■ Braç Vidala; ♦ Murtra; ▲ Sant Climent; ● Reguerons; — Remolar; × Vidala. (s) sampling surface waters; (d) sampling deep water.

sodium (80%). Magnesium (15%) and calcium and potassium make up the rest of the cations. The anions are dominated by chloride (75–90%), followed by bicarbonate (5–17%) and sulphate (3–9%). Remolar surface, Vidala surface, Sant Climent and Murtra lagoons have a high sodium content (50–70%) and less calcium and magnesium (15–25% each). Chloride represents 50–60% and sulphate 8–20%, amongst

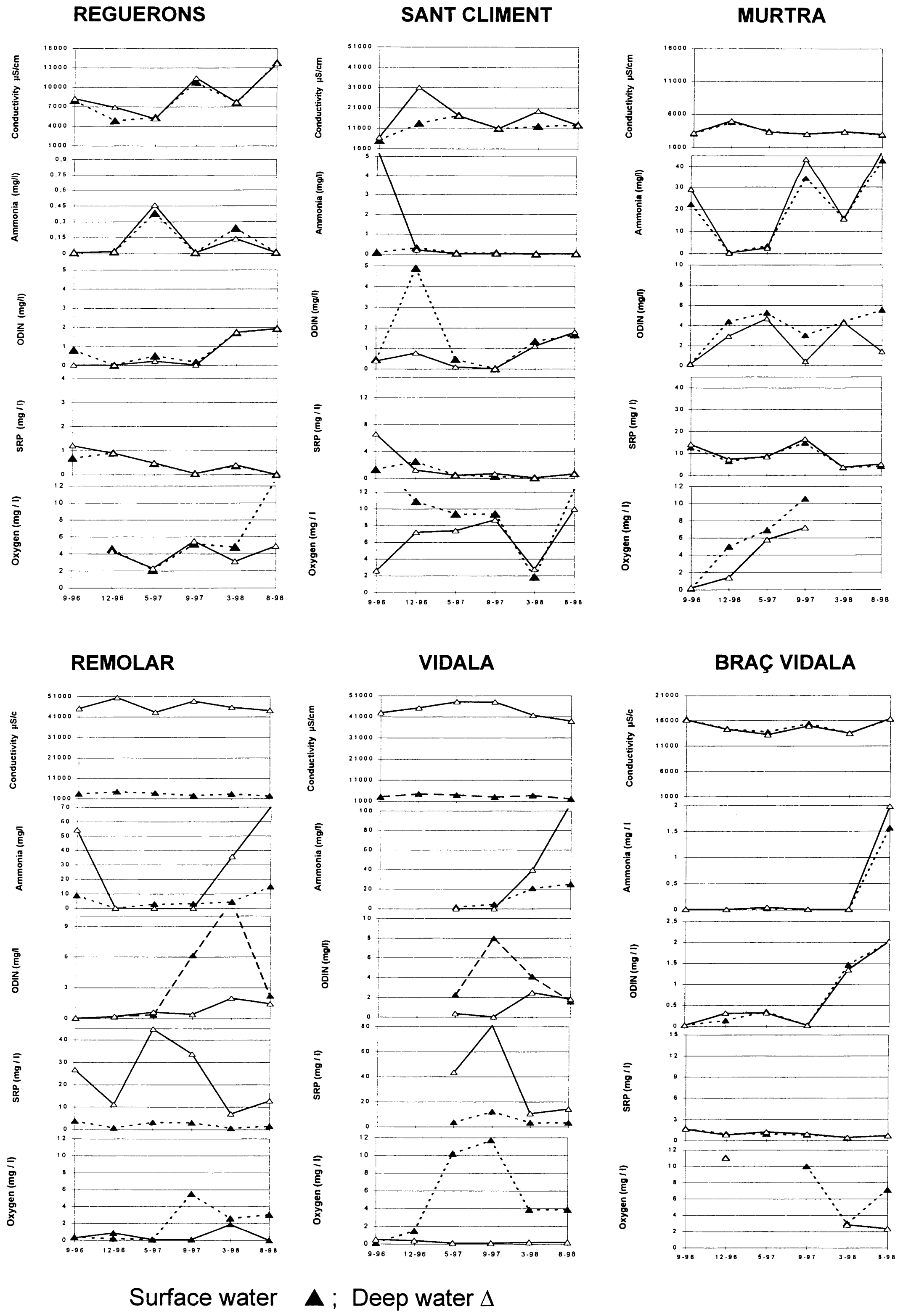

Figure 3. Temporal variations of conductivity, dissolved inorganic nutrients and oxygen in the lagoons of Viladecans.

the anions. The water in the deep part of the water column in Remolar and Vidala contains a very high proportion of chloride (90%), amongst the anions, and sodium (65%), amongst the cations; and the content of magnesium is higher than calcium.

pH

Relatively high values of pH (7.9–8.8) correspond to water masses influenced by seawater under oxygenated conditions, as Braç Vidala and Sant Climent. Lower values (7–7.5) are observed in water masses influenced by freshwater discharges (Reguerons, Murtra and the upper part of Remolar and Vidala). And relatively low values (5.8–5.9) in the anoxic waters, the deep part of the water column of Remolar and Vidala.

Dissolved oxygen

Very low oxygen concentrations (0.1 mg l^{-1}) were observed in the deep part of the water column of Remolar and Vidala throughout the whole period of study. Very low concentrations were observed also on the surface of the water column of these lagoons before September 1997. After this date, the oxygen concentrations observed ranged from 3 to 11.7 mg l^{-1}. This increase was related to the ceasing of freshwater discharges from industrial sources. No temporal pattern of oxygen was observed in the lagoons (Fig. 3).

In the rest of the lagoons the oxygen concentrations ranged from 1 to 13 mg l^{-1}. In general, it was lower in Reguerons, Remolar and Vidala (1–5 mg l^{-1}) than in Sant Climent, Murtra, and Braç Vidala (5–13 mg l^{-1}). However, in the deep part of the water column of these lagoons the oxygen content decreased to 1 mg l^{-1} during a few sampling times.

Nutrients

Relatively low concentrations of dissolved inorganic nitrogen, ammonium plus nitrates plus nitrites (DIN), and SRP, less than 100 and 10 μM, respectively, were observed in Reguerons, Sant Climent and Braç Vidala (Fig. 4). Concentrations higher than 100 μM SRP l^{-1} and about 1000 μM DIN l^{-1} were observed in the deep part of the water column of Remolar and Vidala before 1998. Variable concentrations of DIN and SRP (between 100–6000 μM and 10–175 μM, respectively) were observed in the rest of the water masses. In any case, the highest values were observed in the deep part of the water column of Remolar and Vidala and are mainly due to a high proportion of ammonium compared to other forms of nitrogen.

These data (Fig. 3) are not useful to show seasonal variations of the nutrient concentrations because of the low sampling frequency. An increasing trend of ammonium concentration is observed in Murtra lagoon during the summer months. However, not any consistent seasonal trend is observed in the other lagoons comparing data of the same season corresponding to different years.

Discussion

The ranges of conductivity observed in the lagoons during this study (Fig. 2) shows that three water masses (Murtra, Remolar surface and Vidala surface) are strongly influenced by freshwater because of the relatively low average and standard deviation of conductivity. A second group of lagoons (Reguerons and Braç Vidala) is characterised by brackish water with intermediate values of conductivity. These are lagoons or water bodies with no vertical stratification of the water. In contrast, the lower part of the water column in Remolar and Vidala is quite salty water, accumulated below the freshwater flowing over it. So, a permanent vertical pycnocline is established causing meromixis in these localities. Finally, in Sant Climent a great temporal variation of conductivity is observed from fresh to seawater, which indicates strong inflows from both the continent and the sea.

We hypothesised that seasonal variations of the water characteristics and flows, which are typical in oligotrophic lagoons (Nixon, 1982), are less important than spatial gradients in these lagoons to determine the trophic status of the water masses. It is clear that nutrient inputs are related to freshwater discharges because the lagoons with the highest nutrient concentrations are those with the lowest conductivity, Remolar and Murtra (Figs 5–7). A general view of the nutrient behaviour related to the mixing of water in these lagoons shows two major processes. Dilution of ammonium, nitrate plus nitrite, and SRP takes place during the initial mixing of freshwater in the lagoons, at the lowest part of the conductivity range. Release, increase of ammonium and SRP takes place in the highest part of the conductivity range. This corresponds to water masses in the lower part of the water column of Remolar and Vidala lagoons, which are under anoxic conditions for most of the time. So, the low dissolved oxidised inorganic nitrogen in these waters

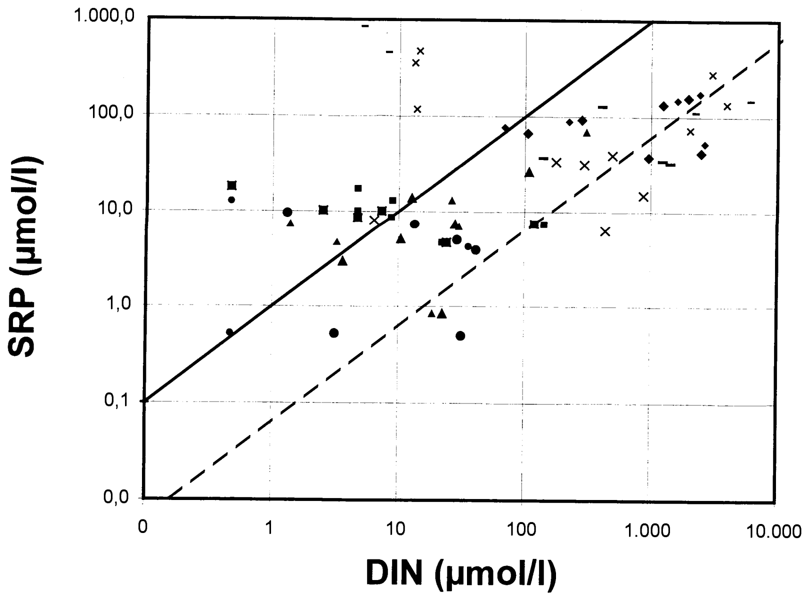

Figure 4. Soluble reactive phosphorus *versus* dissolved inorganic nitrogen in the waters of Viladecans coastal lagoons.

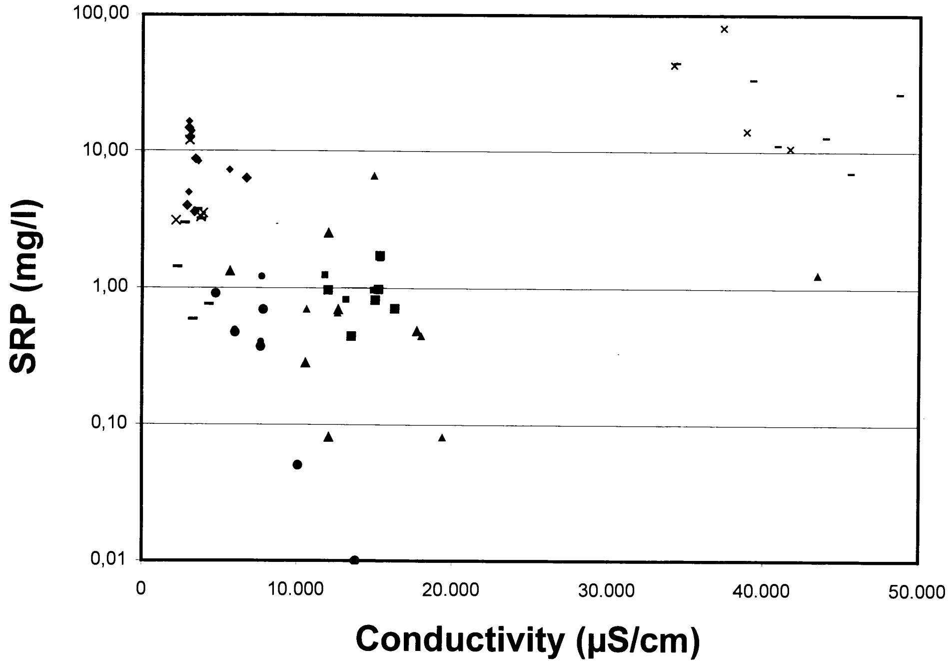

Figure 5. Soluble reactive phosphorus (SRP) related to conductivity in the waters of Viladecans coastal lagoons.

and the high ammonium concentration indicates an intense mineralisation of organic matter and, maybe, denitrification taking place in these sites (Comín et al., 1997).

The range of DIN/SRP ratios observed (0.1–71) shows that a great variety of biogeochemical processes take place in these lagoons (Fig. 4). High DIN/SRP ratios occur in water masses receiving high freshwater discharges (Murtra and the upper part of Remolar and Vidala) and in water masses with high concentrations of ammonium and SRP (the lower part of the water column of Remolar and Vidala after de-

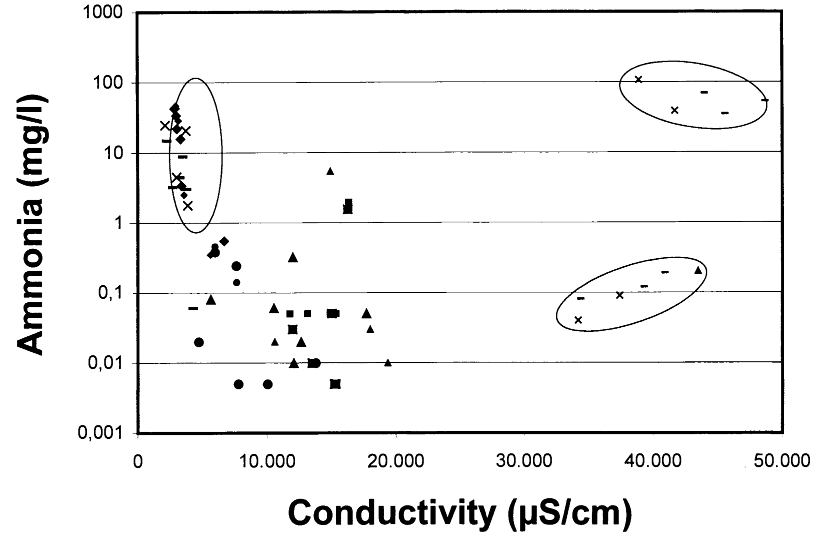

Figure 6. Ammonia (mg l^{-1}) related to water conductivity in the waters of Viladecans coastal lagoons.

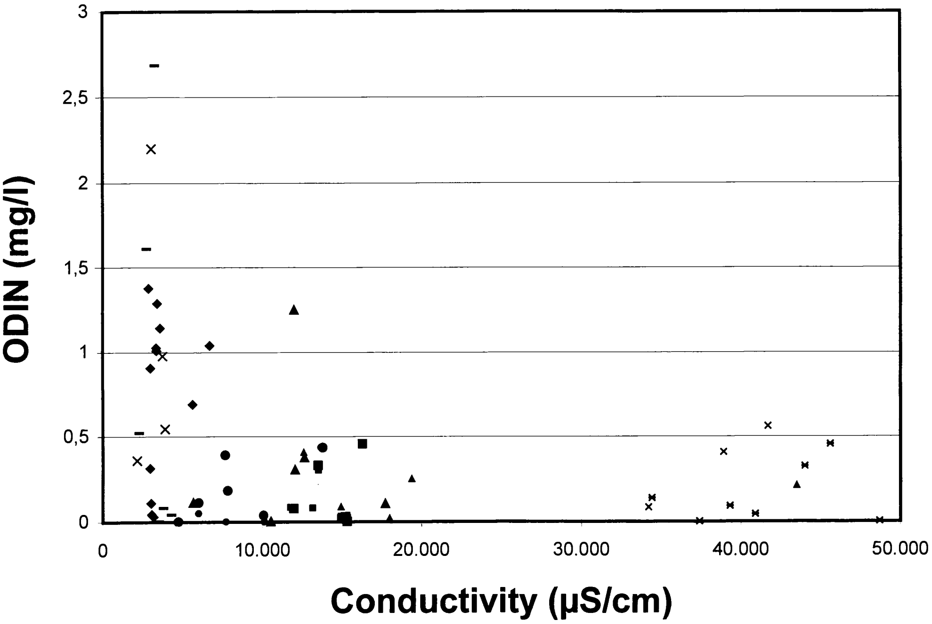

Figure 7. Oxidized dissolved nitrogen (mg l^{-1}) related to water conductivity in the waters of Viladecans coastal lagoons.

creasing organic rich freshwater discharges into the lagoon), which may be originated in the sediment and the lower part of the water column through mineralisation. In contrast, low DIN/SRP ratios correspond to water masses (Reguerons, Braç Vidala, San Climent) receiving water discharges from the aquifer and to those in the lower part of the water column of Remolar and Vidala, at the time high organic rich freshwater discharges take place. Denitrification could be a major biogeochemical process favouring these low DIN/SRP ratios.

Some estimations of the amount of nutrients discharged into the lagoons can be useful to know the level of eutrophication (Vollenweider, 1975; Wetzel, 1981; Ryding & Rast, 1989) (Table 2). This can be estimated for Remolar and Murtra in this study because water flow has been measured in these lagoons. In

Table 2. Reference values for the trophic status of shallow water bodies (after Vollenweider, 1975; Ryding & Rast, 1989; and Wetzel, 1980). Phosphorus (g P m^{-2} yr^{-1}) and nitrogen (g N m^{-2} yr^{-1})

Lagoon		Phosphorus g P m^{-2} yr^{-1}	Nitrogen g N m^{-2} yr^{-1}
Loading	Acceptable	0.07	1
	Excessive	0.13	2
Concentration	Oligotrophic	0.25 μmol	47 μmol
	Eutrophic	2.70 μmol	134 μmol
	Hipereutrophic	31.5 μmol	

the other lagoons, water flow measurement is difficult because non point and quite irregular flows occur.

An amount of 475 m^3 day^{-1} of freshwater has been estimated as the average water flow entering Murtra lagoon through three canals. Taking into account the nitrogen and phosphorus concentrations, it represents 1511 kg N and 603 kg P per year.

In Remolar lagoon, the daily water flow averages 29 560 m^3 (94% of it through one canal only). These waters transport 88 125 kg N and 9505 kg P per year. A high percentage of these inputs is exported to the marine coastal waters (58% N, 52% P). In this lagoon 39 420 kg N and 4562 kg P remain, which represents a net annual loading of 373 g N and 43 g P per m^2 yr^{-1} in Remolar lagoon. In Murtra lagoon, an artificial dike interrupts the flow of water towards the sea. So, no nutrient loss takes place directly to the sea. If all the input is accumulated in the lagoon, it represents 98.8 g N and 39.4 g P per m^2 yr^{-1} in Murtra lagoon. These nutrient discharges are high enough to cause anoxic conditions in the water column of estuaries (Turner & Rabalais, 1999).

Table 2 shows different indicators of trophic state estimated for the studied lagoons. The SRP and DIN concentrations of the water surface in Remolar, Vidala and Murtra indicate that all these lagoons are hypereutrophic. However, the trophic state of Reguerons, Sant Climent and Braç Vidala is lower than in Remolar, Vidala and Murtra. The same is valid for the rest of the water masses sampled. The chlorophyll *a* concentrations are also characteristic of hypereutrophic waters in these lagoons. In fact, the average concentration of chlorophyll *a* is higher in Reguerons and Braç Vidala, the lagoons with lower water turnover, than in the other lagoons. If we apply the equations given by Vollenweider and Wetzel (Vollenweider, 1975; Wetzel, 1981) to estimate the acceptable and excessive phosphorus discharge in a lake, it turns out that Remolar lagoon receives 187 and 330 times higher amounts, respectively, of N and P, than those suggested by Vollenweider (1975) as excessive. In Murtra, they are 25 times higher for N and 150 times higher for P than those, which would be considered as excessive for the metabolism of the lagoon by Wetzel (1981).

These data, both for nutrients and chlorophyll, are much higher than those observed in most Mediterranean coastal lagoons. They are similar to those from water bodies in other parts of the world considered as hypereutrophic, where critical environmental problems have arisen (Table 3). The nutrient content and other characteristics of the lagoons in Viladecans indicate that the trophic state of these lagoons is higher than any other of similar morphological characteristics in Spain.

Another outstanding characteristic is the permanent oxygen deficit observed in those lagoons where water stratification exists. In Remolar and Vidala, very low oxygen concentrations are observed permanently in the lower part of the water column. This is due to the reducing conditions established by organic matter accumulated in the sediments (Hakanson & Janson, 1983). The lack of water turnover due to the vertical salinity gradient contributes to enhance these environmental conditions, which are typical of this type of coastal lagoons (Howard-Williams, 1985). However, in the Viladecans, lagoons the hypereutrophy observed is the result of the extremely high nutrient discharges maintained over many years.

Conclusions

The data presented indicate that the Viladecans lagoons are in a hypereutrophic state, as the result of receiving over a long period of time nutrient discharges

Table 3. Comparative data of phosphorus and nitrogen in Mediterranean coastal lagoons from (1) Vicente (1992); (2) Comín (1994); (3) Quintana et al. (1998); (4) Picot et al. (1990); (5) Comín et al. (1990); (6) Salvat (1997); (7). CCB (1998)

Lagoon	SRP μmol	DIN μmol	Oxygen mg O$_2$ l^{-1}	Chlorophyll-*a* mg m^{-3}
Albufera Valencia (1)	0.47	45.16		486
Massona (2)	2.85	2.69		250
Aiguamolls Empordà (3)	10.45	6.62		21.62
Aiguamolls Empordà (7)	30.81	571.43		
Thau lagoon (4)	1.30	9.65		
Encanyissada (Ebro Delta) (5)	1.56	27.00		2.37
Golf Prat lagoons (Llobregat delta) (6)	3.15	57.7		18.4
Remolar surface (this study)	22.42	393.3	1.87	48
Depth (this study)	239.16	1501	0.3	
B. Vidala (this study)	10	28.89	9.8	284
Vidala surface (this study)	58.29	795.78	5.2	97
Depth (this study)	392.21	2071.67	0.3	
Murtra (this study)	92.67	1243.16	6.45	24
Sant Climent (this study)	13.16	41.92	8.95	313
Reguerons (this study)	4.84	18.71	4.2	223

associated to man-made freshwater inputs higher than their carrying capacity or capacity to metabolise the nitrogen and phosphorus discharges. Remedial measures should include, at least, reducing nutrient discharges into the lagoons and favouring water turnover in the lagoons.

Acknowledgements

To the municipality of Viladecans who encouraged and funded this study.

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Filling of a wetland (Seine estuary, France): natural eutrophication or anthropogenic process? A sedimentological and geochemical study of wetland organic sediments

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Key words: organic phosphate, phosphate fractionation, sediment, wetlands, eutrophication

Abstract

For over a century the Seine estuary has been highly affected by human activities, resulting in a reduction of the surface of wetland habitat. Several ponds of the Vernier Marsh, one of the main wetlands of the estuary, are being filled in by sediment. The St. Aubin canal, the only inlet from the Seine to the wetland, has regularly been opened in summer to maintain a sufficiently high water level in the system. The purpose of this study was to identify the origin of filling: natural evolution of eutrophication or human intervention by the opening of the St. Aubin canal? Sediment cores were sampled at 19 stations randomly distributed in the ponds of the wetland. Granulometric, micro-granulometric and geochemical analyses (ignition loss, C/N/P, chemical fractionation of particulate phosphate) were performed on the top 10 cm of sediment. Moreover nutrient concentrations (ammonium and phosphate) were measured in the interstitial waters using diffusion samplers. The results revealed that sediments sampled from the north are weakly organic (ignition loss: 19%, 15% C, 0.9% N , 40% organic P of total P) whereas those sampled from the southern stations are highly organic (ignition loss: 50–93%, 35% C, 1% N: 59–81% organic P of total P). In summary, sediments accumulating in the north of the ecosystem mainly consist of minerogenic particles from the Seine transported by the St. Aubin canal and thus are a result of the human interference. Interestingly the accumulation of sediments in the southern part is the result of a natural evolution of eutrophication: a peat bog. These sediments are highly organic, enriched with organic particulate phosphate, specifically a humic-acid-bound-phosphorus, a refractory form of phosphorus.

Introduction

Coastal wetlands are favoured sites for the accumulation of fine organo-mineral sediments. They are, therefore, a remarkable sedimentary record both of geological history and of the present evolution of an ecosystem. This, applied to the lower Seine valley, has enabled its Holocene history (the last 10 000 years) to be reconstructed (Huault et al., 1975; Huault & Lefebvre, 1983). The geological history of the Vernier Marsh, the main wetland of the Seine Estuary, has been deduced from paleo-ecological (Huault, 1980) and radiochronological sedimentary markers Huault & Lefebvre (1974). It has thus been shown that the Flandrian transgression has deposited sand and silt with peaty intercalation entailing the formation of an offshore bar isolating the Vernier Marsh from the river.

This wetland has undergone profound changes following developments of the Seine Estuary by Man: channelling the course of the river to improve navigation, draining to increase the areas which can be cultivated etc. These increasing human pressures have damaged the littoral wetlands. The water area in the 'Grand-Mare', the main pond of the Vernier Marsh, has been reduced by half over the last 150 years. Furthermore, this drying up is accompanied by a reduction in flora and fauna biodiversity (Lecomte & Leneveu, 1986).

This ecosystem is closed, the only water inflow comes from a canal ('Saint Aubin Canal') which is directly linked with the Seine. This canal, regularly open in the dry period is used to allow water to be introduced from the Seine to manage the water level. However, the Seine Estuary, like many others, has a zone of maximum turbidity (suspended matter concentration up to 1 g l^{-1}) and it has been shown (Avoine, 1981) that this maximum turbidity zone has migrated upstream. Its present position is located practically at the outlet of the St. Aubin Canal to the Seine. Is the sedimentary impact of the Seine water intakes into the Grand-Mare via the St. Aubin Canal significant and can it explain the filling-in of the system?

Moreover, the sediment is known for its ability to store nutrients (Boström et al., 1988) at a certain period of the year and release them at others (Mesnage & Picot, 1995). This adsorption capacity depends on the amount of iron hydroxides, of calcium carbonate and of organic matter contained in the sediment. Although much is known of the influence of hydroxides on the adsorption since it is quantified by measuring the isotherms of adsorption (Golterman, 1995), less is known of that of organic matter. Fixing depends on the maturity of the organic matter (humic or fulvic acids). This question which has already been raised by Paing et al. (1999) is fundamental to assess the bio-availibility of phosphorus.

In this research, we measure the forms of organic phosphorus by chemical fractionation and not by NMR spectrometry. This choice is based on the composition of the sediment which we have to analyse. In fact, the sediments of the wetlands are very organic and rich in sulphides; these compounds, creating strong interference, reduce the reliability of the identification by ^{31}P NMR.

We have used the C:N:P ratios in surface sediments, a classic tool for characterising organic material. The main functional feature of a wetland is the regeneration of nutrients by biodegradation of the accumulated organic matter. Thus, the interstitial water of the wetland sediment will have strong concentrations of dissolved nutrients. Measuring this dissolved phase allows the exchange flows to be assessed at the water/sediment interface.

Therefore, this research tackles two aspects of studying a wetland, one theoretical (relationship between organic material and particulate phosphorus in sediment) and the other concerning management of the environment (gradual filling-in). The originality of this approach lies in the choice of methods of analysis used, arising from two different fields: sedimentology and geochemistry. Combining the results of these two approaches will allow us to understand the causes of the deterioration of the ecosystem: is it due exclusively to human influence, by opening the canal, or is it simply a natural development of eutrophication?

Materials and methods

Description of site studied

The study was carried out in several ponds of the Vernier Marsh, the main wetland of the Seine Estuary, covering a total surface of 4500 ha. This wetland is located in a former meander of the Seine and has been gradually isolated from the river's influence (see Fig. 1a). Thus, the northern section is a polder while the southern section is a mosaic of natural environments with various ponds. This area is in the process of compression, a consequence of the peat drying out.

The ecosystem studied is composed of three ponds: the 'Grand-Mare' and two adjacent marshes: the 'Crevasse' and the 'Petite-Mare'. The water surface of the 'Grand-Mare' is about 40 ha with a mean depth of 0.5 m. This group of marshes is closed except for a single outlet: a canal (the St. Aubin Canal) which is in direct communication with the Seine. This canal has a tidal valve and a control sluice, but this hydraulic equipment is now old, and therefore, no longer watertight. It has been regularly opened in the dry period to bring water into the system.

Sampling stations, sampling techniques

Nineteen stations randomly distributed on three ponds of the wetland were chosen (see Fig. 1b) and sampled for sediments analyses. For the interstitial water only three stations were sampled (stations no 11, 14, 16, see Fig. 1b).

Undisturbed sediment cores were taken with a manual corer at all stations in March and April 1999 reflecting the spring season. All the analyses have been performed on the top 10 cm. Each sample has been stored in plastic flasks at 4 °C before treatment.

The pore water sampling has been performed with diffusion samplers (Hesslein, 1976). We have used a polysulfone membrane (Gelman) with 0.2 μm pores. Care has been taken to avoid oxygen contamination using bubbling of nitrogen before insertion in the sediment. Samplers were inserted in the sediments and removed after 2 weeks of equilibration time. Removal of pore water samples was done with seringes. The interstitial water samplers were stored at 4 °C during the time of sampling. Phosphate (PRS) and ammonia were analysed.

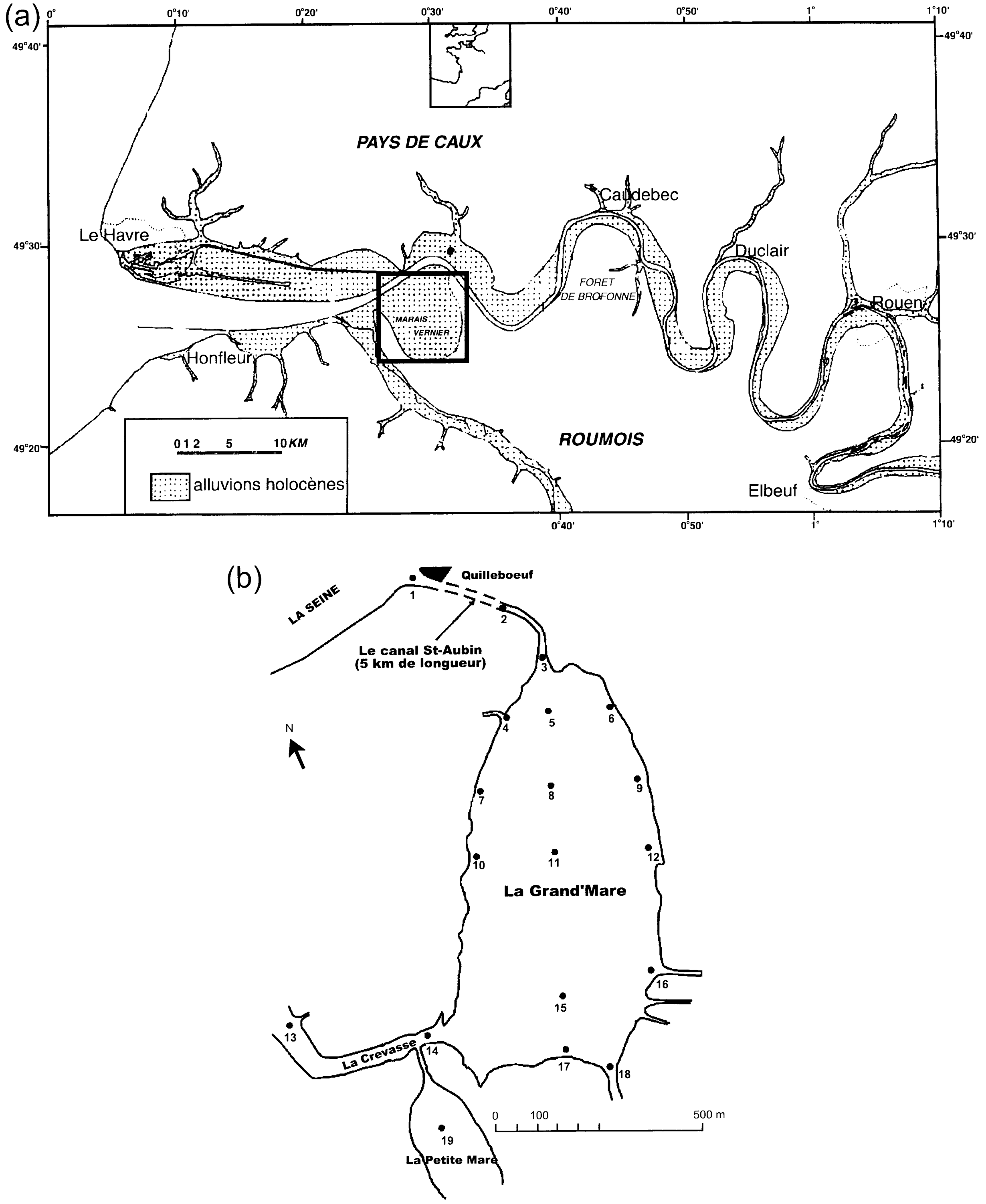

Figure 1. (a) Location of the study site. (b) Location of the sampling stations.

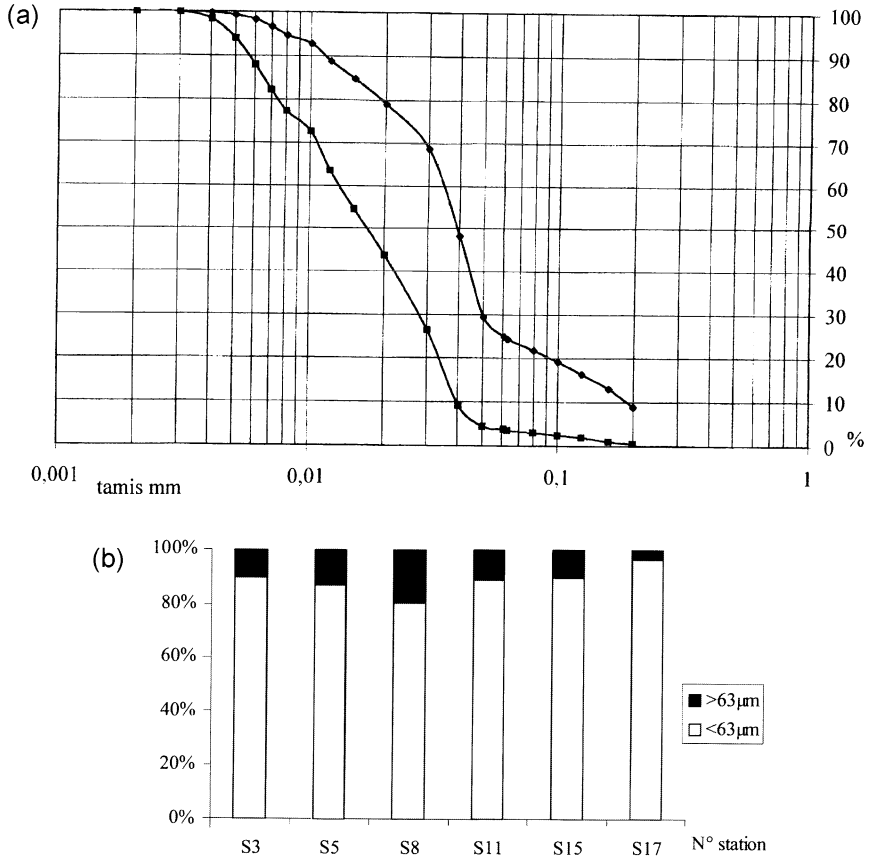

Figure 2. (a) Grain-size distribution of sediments. (b) Grain-size distribution of sediments along a north–south transect.

Chemical extraction

All the analytical methods were performed according to Stainton et al. (1977). Total carbon and nitrogen in sediment were analysed with a C,H,N, autoanalyser after drying the sediment at 105 °C in order to obtain homogeneous sediment samples. Phosphate fractionation was carried out according to De Groot & Golterman (1990) and Golterman et al. (1998). The inorganic fraction phosphate, iron bound-phosphate, Fe(OOH)-P and calcium-bound-phosphate, $CaCO_3$-P, have been extracted with a chelating agent (Na_2 EDTA – 0.1 M with 1% Na-dithionite). The extractions of these two inorganic phosphate fractions were repeated until depletion. Acid Soluble Organic Phosphate (previously ASOP, Golterman & Booman, (1988)) requires a single extraction with H_2SO_4 0.25 M. Then followed an extraction with 2 M NaOH (90 °C) to extract phytate and humic phosphate. All fractionation analyses have been performed in triplicate.

Granulometric analysis

The first stage of the analysis consists in washing the sediment sample with water over a 63 μm sieve in order to separate the fraction >63 μm from that <63 μm. Then the fraction >63 μm, after drying, is treated by classic dry sieving on square mesh screens. The sieves sizes used are as follows: 200, 160, 125, 100, 80 and 63 μm. The fraction <63 μm is analysed by means of a particle counter of the 'Coulter Counter Multisizer' type. Handling consists, first of all, in making the sample conductive by giving it 5% salinity through mixing with a 30% saline solution. The sample is then put into the particle counter. Through a 100 μm diameter aperture the particle-charged solution is pumped up and the difference in potential produced by the particles as a function of their size is measured (Lafite, 1995). A micro-granulometric spectrum in volume is then obtained for 256 channels corresponding to different sizes of particles from 2 to 63 μm.

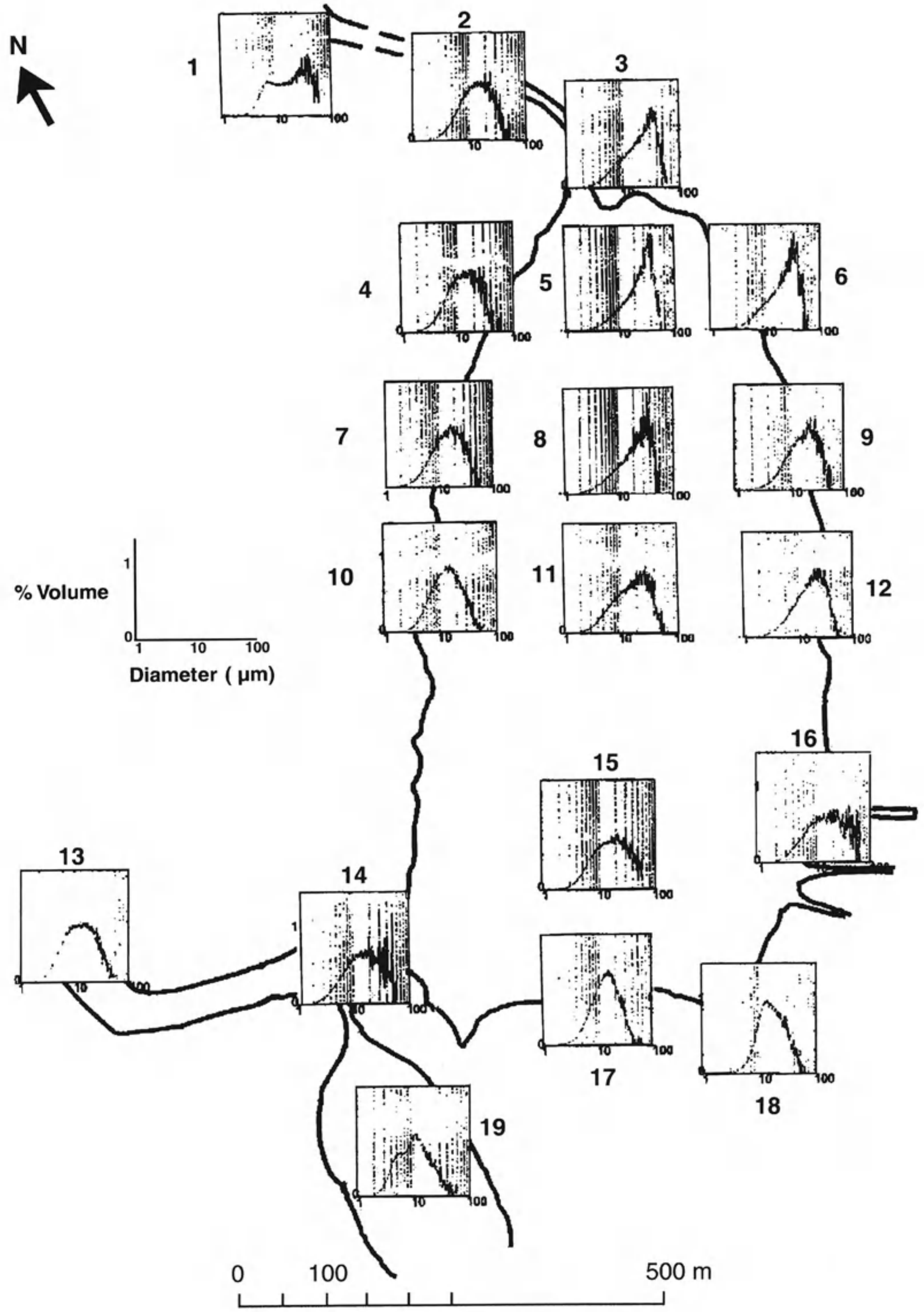

Figure 3. Spatial distribution of the micro grain-size distribution of sediments.

Table 1. Geochemical characteristics of sediments

Stations	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19
Inorganic P. μg/g	–	**588**	**651**	**659**	**411**	**480**	**616**	**613**	**455**	**850**	**860**	**681**	**833**	**522**	**882**	**766**	**778**	**687**	**217**
SD		80	147	96	56	35	147	126	124	96	63	32	134	66	56	63	123	146	235
ASOP μg/g	–	**60**	**60**	**69**	**111**	**142**	**64**	**82**	**115**	**99**	**80**	**73**	**84**	**60**	**114**	**89**	**91**	**107**	**37**
SD		18	31	10	79	109	14	30	84	23	20	4	18	21	13	5	19	15	14
AH-P μg/g	–	**135**	**155**	**122**	**61**	**110**	**147**	**110**	**128**	**190**	**112**	**139**	**289**	**194**	**140**	**160**	**193**	**196**	**255**
SD		35	36	32	8	15	11	83	13	37	80	21	110	31	15	49	28	30	93
AF-P μg/g	–	**338**	**355**	**321**	**167**	**235**	**364**	**269**	**300**	**473**	**321**	**328**	**519**	**398**	**336**	**467**	**497**	**471**	**456**
SD		56	106	22	41	69	85	76	17	100	121	75	65	76	36	139	76	41	102
Ignition Loss %	8	33	27	37	16	18	31	15	27	33	22	39	67	49	18	25	35	41	93
C	6	16	15	19	10	10	17	11	16	18	13	21	35	25	10	15	18	23	46
N	1.7	3	1	2.2	1.3	1.3	1.7	1.6	1.6	1.2	1.6	1.3	1	1	0.5	1	1	2	1
H	0.5	1.8	1.8	2.3	1.5	1.55	2.1	1.2	1.9	2.2	1.4	2.5	4.2	2.8	1.1	1.6	2.2	2.2	5.4

Results

Figure 2a corresponds to the granulometric distribution of all of the sediment samples taken. It appears clearly that the coarse fraction (>200 μm) is extremely small for it only represents 1–9% of the total sediment. On the other hand, the fine fraction (<60 μm) is well represented as it constitutes 70–95% of the total sediment.

Figure 2b shows the results of the total granulometry of the sediments along a North-South transect. The fine fraction represents 80–96% of the total fraction of the sediment and decreases between stations 3–5 to reach the minimum at station 8. This shows that the coarse particles are not deposited in the St. Aubin Canal but are carried along it and deposited at the outlet from the canal into the Grand-Mare.

The results of the micro-granulometric analyses show two main modes of distribution (see Fig. 3): one concentrated around 10–20 μm, the other concentrated around 35–40 μm. Moreover, these modes are spatially distributed over the whole ecosystem. The sediments of station 1, located at Quillebeuf at the Seine (see Fig. 1b), as well as those of the north of the Grand-Mare (stations 3, 5, 6, 8) have a micro-granulometric distribution concentrated around 35–40 μm, while those of the south and west edge have a distribution concentrated around 10–20 μm.

In order to characterise the nature of these 2 micro-granulometric modes, we treated the sediments of three stations (3, 11, 15) with hydrogen peroxide and then did a μ-granulometric analysis (see Fig. 4). The sediments treated with hydrogen peroxide have a mode concentrated around 35–40 μm, whereas the untreated sediments have a distribution concentrated around 10–20 μm (see Fig. 4). Therefore, the mode concentrated around 35–40 μm characterises mineral particles while that concentrated around 10–20 μm characterises organic particles.

Table 1 groups together the results of the geochemistry of the sediments and Figure 5 presents, the results of particulate phosphate, spatially. In the light of the results listed in Table 1, it is possible to isolate three different types of sediment from their composition ratios of organic material (loss on ignition), carbon and nitrogen. The sediments of the Seine (station 1) have figures of 8%, 6% and 1.7%, respectively, for the loss on ignition, carbon and nitrogen. The average of the concentrations worked out for the sediments of the northern and central section of the Grand-Mare (stations 3, 5, 8, 11, 15) gives figures of 19.2% for loss on ignition, 15% for carbon and 0.86% for nitrogen; then 50–93% loss on ignition, 35% carbon, 1% nitrogen for the south area (stations 13, 14, 19). Therefore, the geochemistry of the sediments confirms the spatial arrangement of the ecosystem already observed with

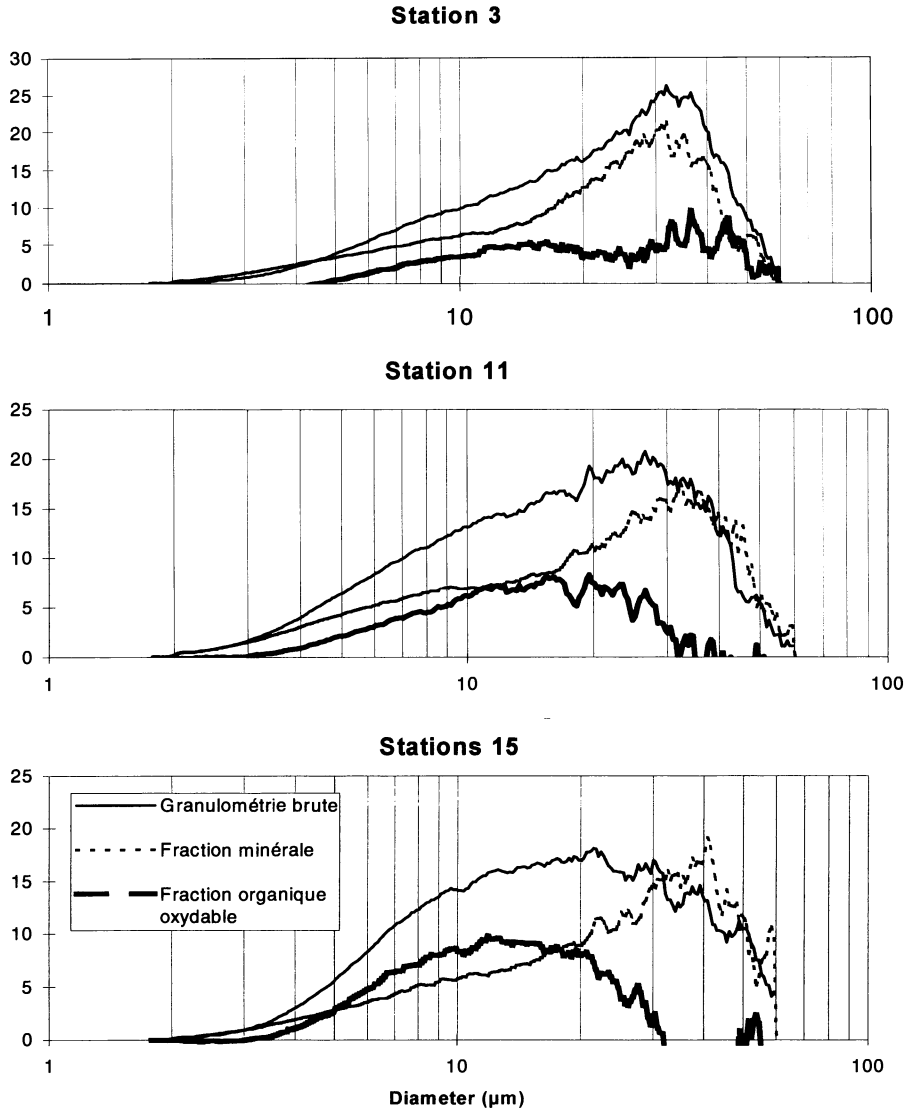

Figure 4. Micro grain size distribution of sediments after peroxide treatment.

the results of the μ-granulometry, i.e. a northern and central area composed of mineral sediments compared with a southern area where the sediments are organic.

The results of fractioning the phosphorus (Fig. 5) allow more detailed insight into the geochemical composition of the sediments. The organic phosphorus is the form of particulate phosphorus dominant in the sediments studied, with concentrations varying from 43% (station 11) to 81% (station 19) of the total phosphorus. Inorganic phosphorus concentrations represent overall 50% of the total phosphorus for all the stations except for the stations located in the south of the ecosystem (stations 13, 14, 17, 18, 19). The inorganic phosphorus concentrations vary from 19% (station 19) to 57% of the total phosphorus; it is at station 19 that these concentrations are minimal with a figure <20%, so at this station the organic phosphorus is in the majority.

Further, the fractionation of organic phosphorus enables three main forms to be identified: ASOP (Acid soluble organic phosphate) AF-P (fulvic acid bound P) and AH-P (humic acid bound P) showing significant variations in concentration.

The ASOP concentrations vary from 3% (station 19) to 14% (station 1) of total phosphorus. The sediments of the northern part of the ecosystem (the

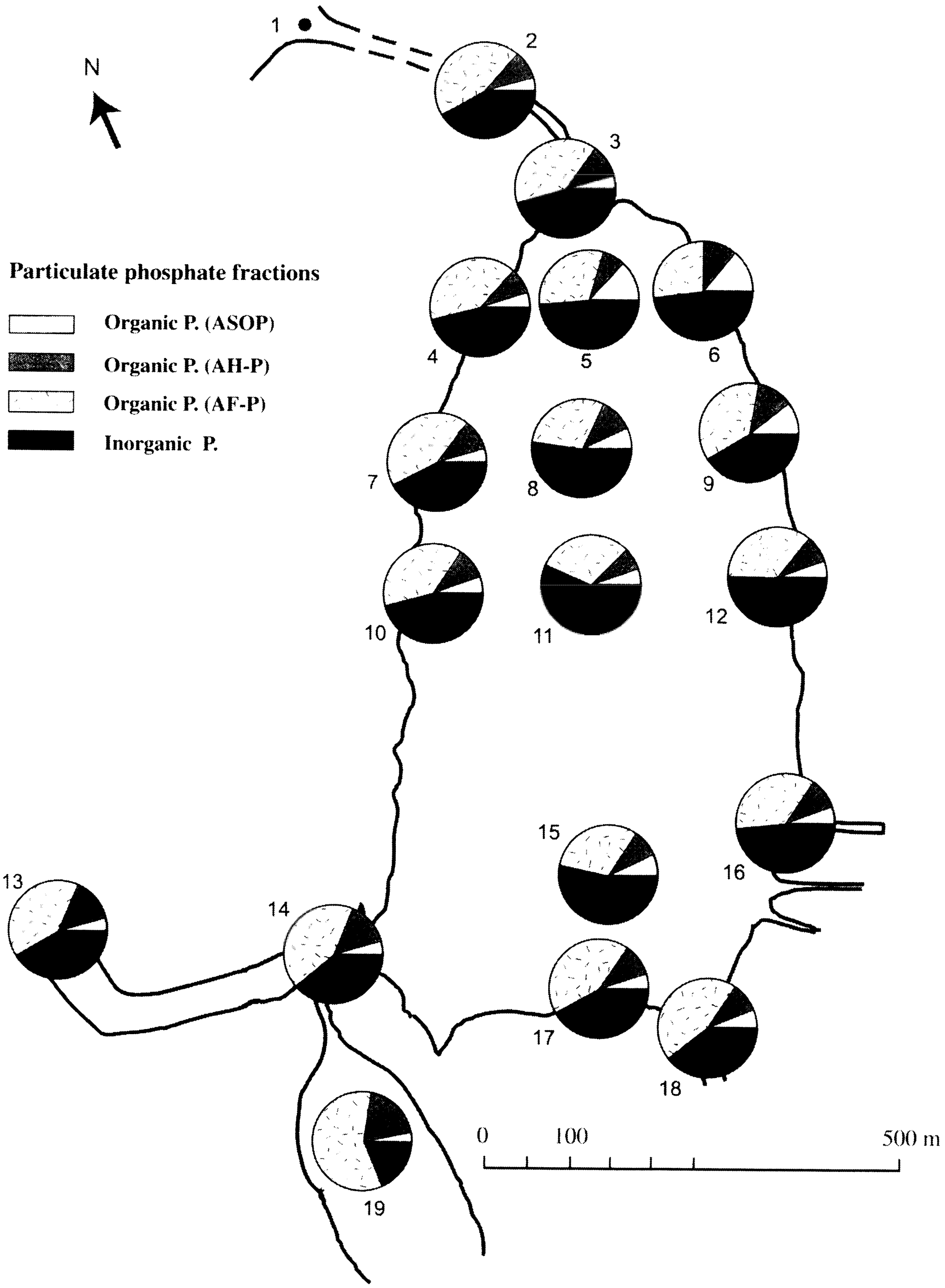

Figure 5. Spatial distribution of the particulate phosphate fractions in sediments.

Grand-Mare) have the strongest concentrations of ASOP, higher than the concentrations measured in the sediments of the stations in the south (Crevasse and Petite-Mare).

This North–South zoning of the ecosystem also exists for the forms of phosphorus linked to humic acids (AH-P), but it is reversed, i.e. it is the stations in the south of the ecosystem which have the strongest concentrations. The concentrations vary from 7 to 11% for the sediments of the Grand-Mare and from 14% to 19% for the other two ponds. As to the concentrations of phosphorus linked to the fulvic acids (AF-P), they vary from 19% to 40% of the total phosphorus but do not show any spatial variation (see Fig. 5).

The results for the chemistry of the interstitial water are shown in Figure 6. For station 11, a gradient of concentration of phosphate and ammonium exists and is maintained in the first 30 cm of sediment. The maximum concentrations of PO_4 reach 4 mg l^{-1} and 3 mg l^{-1} for NH_4. At station 16, a gradient of concentration also exists but disappears in the first 15 cm of sediment. The concentrations of PO_4 also reach figures of 3 mg l^{-1}; on the other hand, the concentrations of NH_4 are much lower and do not exceed 1 mg l^{-1}. The rate of the concentration profile of NH_4 and PO_4 at station 14 is very different compared with the other stations. This profile does not show any clear gradient of concentration and the levels of concentration remain high over the full depth of sediment studied.

They are in the region of 6 mg l^{-1} of PO_4 and 35 mg l^{-1} of NH_4. These profiles enable the difference in nutrient level to be shown between station 14 (the 'Crevasse') and the other two stations studied, and confirm the fact that the southern part of the ecosystem is very rich in organic material.

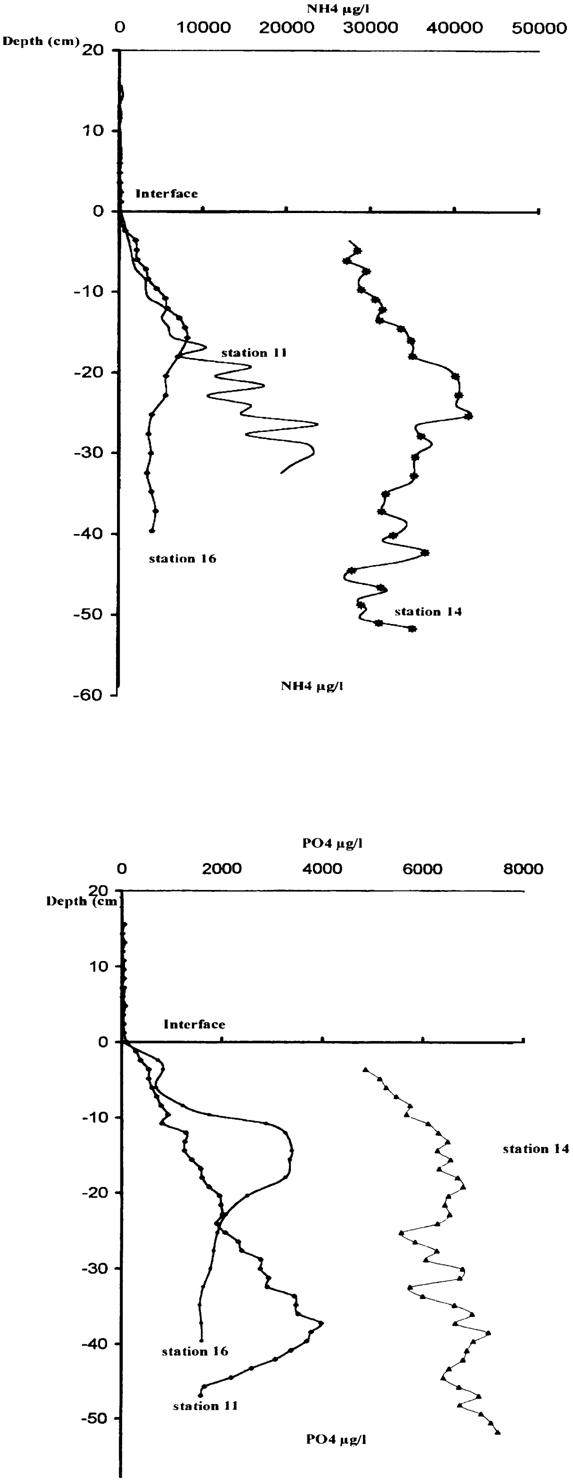

Figure 6. Interstitial water chemistry: pore water profiles for ammonia and phosphate.

Discussion

A number of granulometric arguments point in the same direction to show that there is a real input of coarse particles coming from the Seine, carried along the canal into the Grand-Mare: the results of the overall granulometry show the increase in coarse particles along a North–South transect (see Fig. 2b) and the results of the granulometric modes show the predominance of coarse mineral particles to the north and centre of the system (see Fig. 3).

The sedimentary impact of the Seine is undeniable, carrying sediments of mineral nature with low carbon and nitrogen contents. These fairly low mineral addi-

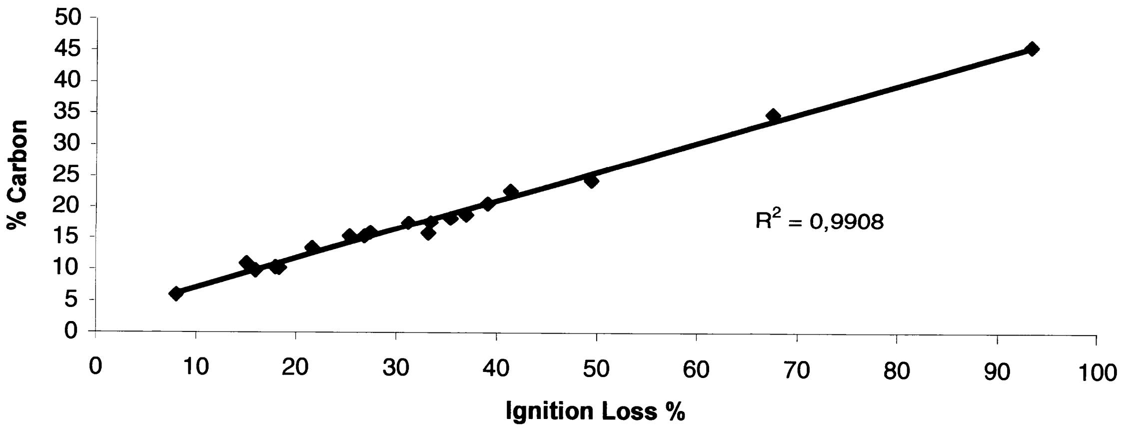

Figure 7. Relation between % C versus organic matter (Ignition Loss).

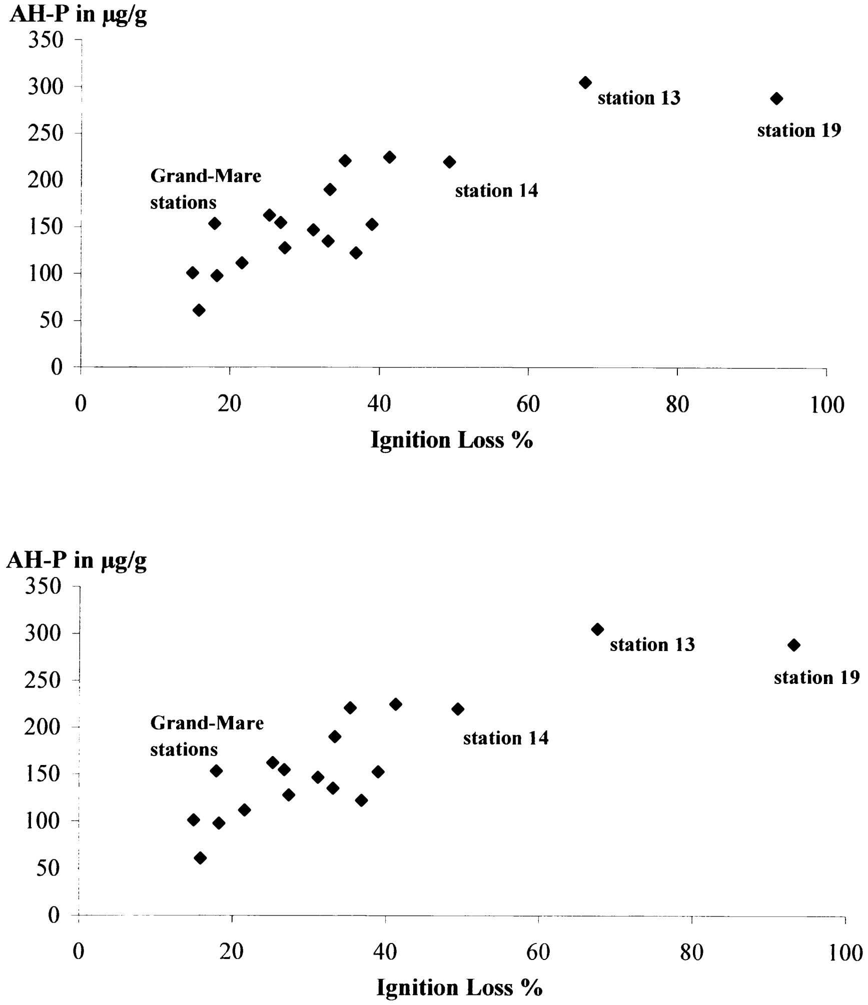

Figure 8. Relation between particulate organic phosphate (ASOP, AH-P) and organic matter.

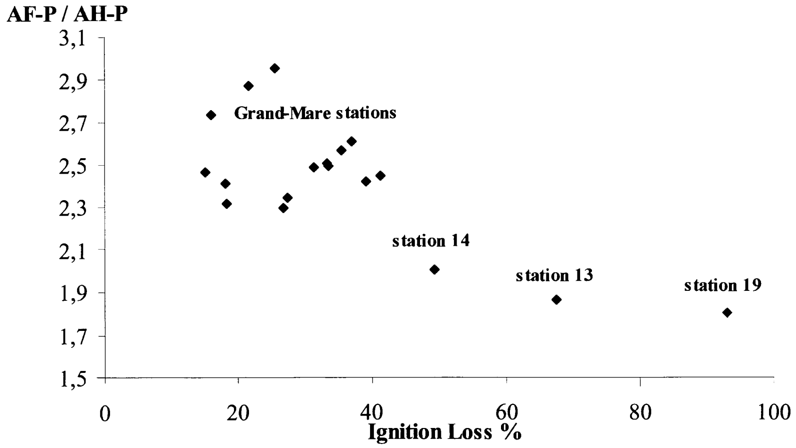

Figure 9. Correlation between organic matter maturity and phosphate fractions.

tions make the C:N ratios drop. Thus, the accelerated deterioration of the ecosystem is explained by these intakes of water highly loaded with material in suspension. Environmental management for maintaining the water levels by opening the St. Aubin Canal must be reviewed in order to limit the particulate additions and minimise the filling-in of the Grand-Mare.

The variation in the C:N ratios within the ecosystem is explained by the diversity of the vegetation on the banks of the marshes. Thus, the nature of this vegetation (reeds, oak trees, birch trees etc.) will influence the C:N ratios of the sediments. The average figure for the C:N ratios for the stations on the edge of the Grand-Mare ecosystem (stations 4, 7, 10, 6, 9, 12, 16, 17, 18) is 14. It is explained by the presence of a reed bed encircling the Grand-Mare with the common reed (*Pragmites communis*) as the dominant form, its C:N ratio being 12 (Tezuka, 1990). The sediments of the northern part of the ecosystem (stations 2, 5, 8) have a much lower C:N ratio due to input of sediment from the Seine with a C:N ratio of 3 (station 1). Therefore, the input of sediment coming from the Seine bring about a drop in the C:N ratios for the sediments in the northern part of the Grand-Mare ecosystem. The very strong figures of the C:N ratio for the 3 stations of the adjacent ponds (13, 14, 19) are explained by the accumulation of organic material of peaty origin. It has been shown in particular that particulate organic phosphorus (AH-P) is present in strong concentrations, formed by the presence of this resistant organic material (peat).

The difference in the nitrogen contents of the sediments of the Grand-Mare ecosystem in comparison with those of the sediments of the coastal lagoons is mainly due to the source of the additions of nitrogen. In fact, in the Grand-Mare, the input of nitrogen have come from decomposition of the heavily nitrogen-charged reeds established on the banks (N:P of the reed: 62, Tezuka, 1990). There is no input of nitrogen, or very little, from phytoplankton (N:P: 5–9, Hecky & Kihlam, 1988) nor from macro-algae, as may be the case in Mediterranean coastal lagoons for example.

Further, the sediments of the Grand-Mare are overall enriched with nutrient elements and with organic material compared with other ecosystems such as the lagoons which are also littoral wetlands. For this type of ecosystem, the figures for carbon do not exceed 10% and those of the C:N are 22 maximum (Gomez-Hernandez, 1996).

Figures 7, 8 and 9 allow us to discuss the role of the humic substances on the biogeochemistry of particulate phosphorus. Humic acids are resistant organic compounds which come from the evolution of fulvic acids. As the phosphorus, which associates with this organic material, will form AH-P, it forms resistant particulate organic phosphorus which will accumulate in the sediments. Thus, these forms of phosphorus will be difficult to mineralise and will not, therefore, be bioavailable.

Figure 8 sets the forms of organic phosphorus (ASOP or AH-P) and the organic matter (loss on ignition) in relation to each other. Its objective is not to show the obvious relation between loss on ignition and the form of organic phosphorus, but to show that three plots are always isolated from the overall cloud of plots. These three plots correspond to the 3 stations located in the two adjacent ponds (Crevasse and Petite

Mare). There really is an accumulation of particulate organic phosphorus in these adjacent ponds.

The variation in the AF-P:AH-P ratio as a function of the loss on ignition at each of the stations (see Fig. 9) allows us to estimate the influence of the state of degradation of the organic material on the formation of particulate organic phosphorus. The more advanced the degradation of the organic material (humic acids), the more the forms of particulate phosphorus associated with the humic acids (AH-P) will be present in the sediments. The lowest AF-P:AH-P ratios are found at the most organic stations, i.e. in the adjacent pools.

Thus, it is shown that the state of degradation of the organic material influences the fixing of the phosphorus in the sediments. In fact, the phosphorus which fixes on to the 'fresh' organic material gives compounds of the ASOP type while the phosphorus which associates with more stable forms of organic material gives AH-P forms. ASOP is a form of organic phosphorus which is easily soluble in acid. In general, this fraction represents only a small part of the particulate phosphorus (1–5%) and does not show any variation with time (on the seasonal scale) in the sediments (Mesnage & Picot, 1995). In the sediments of the Grand-Mare, this form of organic phosphorus is represented somewhat more and can reach a proportion of 14% (station 1).

The spatial variation in the distribution of the two forms of particulate organic phosphorus is explained by the communication between the ecosystem and the Seine (via the canal). The rather higher concentrations of ASOP in the sediments in the north of the ecosystem are interpreted as input of organic material which have come from micro-algae (phytoplankton). This 'fresh' organic material from the waters of the Seine is added through the St. Aubin Canal up to and into the Grand-Mare. It must be said that even if the canal is no longer open, leakage occurs at the hydraulic equipment on the canal (lock, tidal valve), which can explain the regular flows of water from the Seine into the Grand-Mare ecosystem.

Conclusion

All of the results (granulometry, C/N/P, phosphate fractionation etc.) allow it to be concluded that this ecosystem is sectored: in the north section, mineral particles which have come from the Seine and been carried by the St. Aubin Canal, have accumulated. The south section, on the other hand, consists of organic sediment which is very rich in particulate organic phosphorus specifically connected to humic acids (AH-P). Thus, the gradual filling in of this ecosystem is explained to the north by the opening of the canal and is therefore of human origin, whereas in the south it is essentially a question of natural evolution of eutrophication towards a peat bog.

Further, the variation in the AF-P:AH-P ratio as a function of the loss on ignition at each of the stations has allowed us to estimate the influence of the degradation of the organic material on the fixing of the phosphorus, either on the humic acids or on the fulvic acids. The lowest AF-P:AH-P ratios are found at the most organic stations, i.e. in the adjacent ponds.

Furthermore, it shows that the maturity of the organic material influences the fixing of the phosphorus in the sediments. The index of degradation (diagenesis) of the organic material by using the methods of molecular geochemistry (measurements of sugars etc.) remains to be measured.

Acknowledgements

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The effects of macroalgal cover on the spatial distribution of macrobenthic invertebrates: the effect of macroalgal morphology

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Key words: macroalgal mats, sandflats, macrofauna, sediment variables

Abstract

The growth of green macro-algae in response to nutrient inputs is a common phenomenon in marine estuaries and sheltered bays. While the ecological effects of the growth of the most commonly occurring macroalgal taxa (*Enteromorpha*, *Chaetomorpha*, *Ulva*, *Cladophora*) have been well studied, the effects of a morphologically very different species, *Vaucheria subsimplex*, have not been investigated. This study investigated the ecological effects of the establishment of *V. subsimplex* on a relatively exposed intertidal sandflat, Drum Sands, Firth of Forth, Scotland. Because of the spatially heterogeneous development of the weed, the short term (4 weeks) and long term (20 weeks) effects of the weed could be studied using a survey approach in which the weed-affected and weed-free plots were interspersed. After 4 weeks, *V. subsimplex* significantly increased the mean number of individuals and diversity of the macrofauna, eight of the ten most abundant species showed significant increases in abundance compared to weed-free areas. After 20 weeks, mean number of species and individuals were significantly higher under weed patches, while species diversity was reduced due to the numerical dominance of *Pygospio elegans* (Claparède). The weed, therefore, had an enriching effect on the macrofaunal communities on Drum Sands. The increased numbers of *P. elegans*, the numerical dominant infaunal species on Drum Sands, resulted mainly from enhanced larval recruitment to weed-affected areas. The effects of *V. subsimplex* on sediment characteristics were similar to those reported for other macroalgal taxa, i.e., increased water, organic and silt/clay contents, medium particle size and sorting coefficients, and reduced redox potentials. The results from this study are compared to those for other, morphologically different macroalgal species, with particular reference to an *Enteromorpha*-implanted experiment on the same sandflat. Since the general effect of such macroalgal taxa on macrofaunal communities is a detrimental one, the present study supports the contention that macroalgal morphology is an important feature in algal–faunal interactions.

Introduction

Excessive growth of green macro-algae in response to nutrient inputs is a common phenomenon in marine estuaries and sheltered bays (Pihl et al., 1999). In temperate regions, the growth of mat-forming algae occurs mainly in the spring and is capable of persisting at high maintained densities throughout the summer before disappearing in the late autumn (Hull, 1987). The ecological effects of such macroalgal growth have been well documented (Perkins & Abbott, 1972; Fahy et al., 1975; Wharfe, 1977; Nicholls et al., 1981; Soulsby et al., 1982, 1985; Reise, 1983, 1985; Tubbs & Tubbs, 1983; Thrush, 1986; Hull, 1987, 1988; Ólafsson, 1988; Sundback et al., 1990; Everett, 1991, 1994; Raffaelli et al., 1991, 1999; Bonsdorff, 1992; Bolam et al., 2000; Raffaelli, 2000). Generally, macroalgal mats, usually of the genera *Enteromorpha*, *Chaetomorpha*, *Ulva* or *Cladophora*, cause the underlying sediments to become more reducing, often leading to anoxia and the accumulation of toxic hydrogen sulphide (Wharfe, 1977; Reise, 1985). Most macrofaunal invertebrates are unable to tolerate these physico-chemical conditions (Reise, 1985; Raffaelli et al., 1991; Raffaelli, 2000) and the resulting communities become dominated by opportunistic species (Thrush, 1986) resembling those occurring in areas of high organic loading (Pearson & Rosenberg, 1978). The resulting changes in infaunal invertebrates have an indirect effect on the numbers of predators, notably wading birds (Fahy et al., 1975; Nicholls et al., 1981; Soulsby et al., 1982; Tubbs & Tubbs, 1983; Raffaelli, 2000) and fish (Perkins & Abbott, 1972; Raffaelli, 2000) that these areas are able to support.

Generalisations concerning the effects of weed cover on soft-sediment invertebrate assemblages are very difficult and their effects still remain unclear (Everett, 1994). Differences in the nature of the environment, the genera of the algal species, whether the alga is attached or floating and differences in the weed biomass all have different effects upon the fauna (Raffaelli et al., 1999). This makes direct comparisons between studies very difficult. Previous studies investigating the effects of weed mats on the sediments and associated fauna have been either descriptive surveys (e.g. Perkins & Abbott, 1972; Fahy et al., 1975) or controlled, manipulated experiments (e.g. Woodin, 1977; Reise, 1983; Hull, 1987, 1988; Everett, 1994; Bolam et al., 2000). Surveys have involved comparing the invertebrate communities of areas where algal mats are present to those where they are absent, usually in another part of the same estuary or sandflat (e.g. Nicholls et al., 1981). However, Lowthion et al. (1985) suggested that the presence of algal mats may be influenced by factors other than nutrient availability and that these factors may affect the infaunal communities independent to the effects of the weed. Consequently, because of their large-scale separation, weed-affected and weed-free areas which have been used in observational studies may have been completely different environments and, therefore, not comparable.

In this study, we investigate for the first time the effects of the establishment of *Vaucheria subsimplex* on an intertidal sandflat, Drum Sands, Scotland. The physical nature of this algal species is very different from those previously studied, and therefore, this study allowed us to test whether the effects of *V. subsimplex* on the benthos were similar to those reported for macroalgal species with a very different morphology. We compare the effects of *V. subsimplex* with those obtained from an *Enteromorpha*-implantation experiment on the same sandflat (Bolam et al., 2000). The spatially heterogeneous development of the weed cover in this study allowed us to overcome the inherent problem of surveys: weed-affected and weed-free areas were interspersed. Furthermore, we investigate the mechanisms of algal cover–invertebrate interactions with reference to the most dominant infaunal species on Drum Sands, the spionid polychaete *Pygospio elegans*.

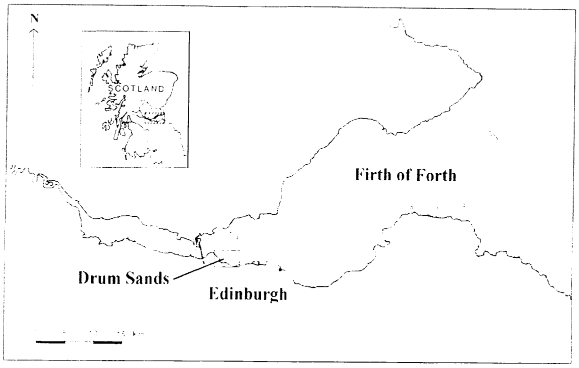

Figure 1. Map of the Firth of Forth, showing the position of Drum Sands.

Methods

Study site

Drum Sands is located in the Firth of Forth on the east coast of Scotland (Fig. 1). A full account of the general ecology and environment of the area can be found in Webb & Metcalfe (1987) and Bolam (1999). Generally, the biological and chemical characteristics of this sandflat are good with no detectable effects from a nearby oil exporting terminal or treated ballast water outfall. Dominating macrofauna are the polychaete *P. elegans* and the bivalves *Cerastoderma edule* (L.) and *Macoma balthica* (L.). Macroalga (*Enteromorpha prolifera* (Müller) and *Vaucheria subsimplex*) have established on this site during recent years (Bolam, 1999). Drum Sands is relatively flat and the majority lies below mid-tide level. The sediments within the present study area are moderately sorted with a median particle size of 2.6ϕ, loss on ignition 3–5%, and with no obvious environmental gradient (Bolam, 1999).

Sample design

During late August 1997, patches (1–2 m^2) of green algae (*V. subsimplex*) developed on Drum Sands. These covered roughly 30–35% of the sediments within the area and approximately 100% within patches. On the 18th September (approximately 4

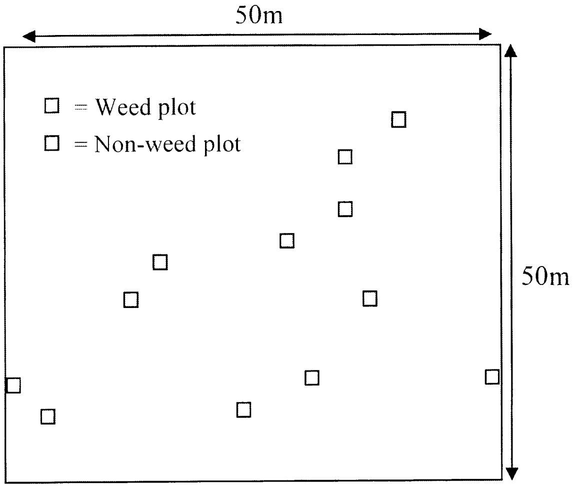

Figure 2. Random layout of the weed and non-weed plots.

weeks after the weed first established), six 'weed' plots and six 'non-weed' plots, 1 m^2 each, were randomly established within a selected area (50×50 m) of the area affected by macroalgal cover (Fig. 2). This was achieved by choosing the nearest weed-affected and weed-free area to the randomly generated co-ordinates for the weed and non-weed plots respectively. The assumption that the weed had no effect on the characteristics of the non-weed plots was made. This assumption seemed reasonable with respect to hydrodynamic effects at least since *V. subsimplex* forms very short filaments, and therefore, has a very localised hydrodynamic effect.

The sediments could not be sampled prior to algal development since weed patches had to establish before the two plot types could be positioned. Therefore, there is the assumption that the faunal and sedimentary characteristics in the weed plots were initially identical to the non-weeded controls, i.e. that algal establishment within the area was random. This assumption seemed reasonable with respect to the numerically dominant species, *P. elegans* (Bolam, 1999).

The plots were sampled for their invertebrate fauna and sedimentary characteristics (hereafter referred to as '4 weeks' samples). The fauna was sampled by taking three cores (each 6×6 cm in area, 10 cm depth), located within each plot by random numbers. The fine filaments of *V. subsimplex* are found in close association with the sediment and so could not be removed when sampling the macrofauna. Contrary to mat forming benthic macro-algae, *V. subsimplex* filaments protrude less than 1 cm above the sediment surface and thus do not cover it.

Cores were sieved on a 500 μm mesh sieve and preserved using neutralised, saline formaldehyde solution (10%) with 0.01% Rose Bengal, and the organisms identified to the lowest possible taxonomic level. The three cores were then pooled to provide a single value for each plot. The sediment samples were taken within each plot by three randomly located cores (2.4 cm internal diameter, 3 cm depth) and the data pooled as above. As much as possible, weed was carefully removed from the sediment before the cores were inserted to ensure that differences in organic content were not due to weed biomass. Water content was determined by weight loss on drying at 100 °C, organic content by weight loss on ignition at 480 °C for 4 h and particle size analysis was carried out after Holme & McIntyre (1984). Percentage silt/clay (<63 μm), median particle size (Md ϕ) and sorting coefficient (Inclusive Graphic Standard Deviation) were derived from graphical plots of the particle size distributions. Redox potential was measured at 1, 2 and 4 cm depths after Pearson & Stanley (1979) using a RL100 meter (Russel Ltd.). Sampling was repeated on the 8th January, 1998, 20 weeks after algal establishment (hereafter referred to as '20 weeks' samples). The weed had been slowly disappearing since mid-November and by this time had almost completely disappeared.

One random sample (6×6 cm) was taken from each plot for weed biomass determination after 4 weeks and 20 weeks. The weed was washed to remove any fauna or sediment and dried at 80 °C for 24 h to constant weight (Everett, 1991).

Size measurements of all *P. elegans* individuals were measured from the weed and non-weed plots for both the 4 weeks and 20 weeks samples. This was carried out by measuring the width of the 5th setiger using a dissecting microscope (×40 magnification) fitted with an eye-piece micrometer.

Data analyses

Only those species with a mean abundance of three or more per core were included in the analyses. All data were checked for normality using the Anderson–Darling test and homogeneity of variance using the Bartlett test. Data not meeting these criteria were transformed appropriately (see Zar, 1984) and again checked for normality and heteroscedasticity. *T*-tests were then carried out on either the raw or transformed data to test for differences between the weed and non-weed plots. A Mann–Whitney *U*-test was

Table 1. Mean number of species and individuals (±S.E. Mean, n=6) and Shannon–Wiener diversity index values for the non-weed and weed plots for 4 and 20 weeks results. Significant differences are indicated by p values while N.S. represents non-significance (two-sample t-test except for * where significant differences were assessed by Hutcheson's modified t-test)

	Mean no. species	p	Mean no. individuals	p	Shannon–Wiener	p
4 weeks						
Non-weed	10.6 (±0.61)	N.S.	76.3 (±9.0)	**<0.001**	0.563	* **<0.01**
Weed	10.6 (±0.56)		192.8 (±8.2)		0.733	
20 weeks						
Non-weed	8.5 (±0.34)	**<0.001**	39.7 (±7.9)	**<0.001**	0.789	* **<0.01**
Weed	11.0 (±0.37)		301.2 (±30.0)		0.516	

carried out on any variable not satisfying the parametric assumptions even after transformation. *P. elegans* size distributions were compared between weed and non-weed plots by Kolmogorov–Smirnov tests.

Results

Macrofauna

The invertebrate abundances for the 4 weeks and 20 weeks samples are shown in Figure 3(i) and (ii), respectively. Table 1 shows the mean number of species, individuals and Shannon–Wiener diversity index values for the two plot types. After 4 weeks of algal cover, the total number of individuals and diversity significantly increased in the weed plots while there was no change in the mean number of species. Furthermore, there was no change in the total number of species: 17 species were obtained from the six replicates for both the weed plots and the non-weed plots, ten being sufficiently abundant for statistical analysis. Eight of these showed significant differences in abundance between the two plot types, all being more abundant in the weed plots compared to the non-weed plots. These were the polychaetes *P. elegans*, *Capitella capitata* (Fabricius), *Streblospio benedicti* (Webster), *Polydora cornuta* (Bosc) and *Eteone* cf. *flava* (Fabricius), the bivalve *M. balthica*, the amphipod *Corophium volutator* (Pallas) and the nudibranch *Doto* sp., the latter two being only sampled in the weed plots. *C. edule* (L.) and *Anaitides mucosa* (Oersted) showed no significant change in abundance with algal cover and no species was significantly more abundant in the non-weed plots.

After 20 weeks, the mean number of species and individuals in the weed plots were still significantly higher than in the non-weed plots, however, due to the dominance of *P. elegans* under algal cover, the diversity significantly decreased. A total of 16 species were sampled from the non-weed plots and 15 species from the weed plots, ten species being sufficiently abundant for statistical analysis. Of these ten, six species (*P. elegans*, *C. capitata*, *A. mucosa*, *S. benedicti*, *P. cornuta* and *C. volutator*) showed significant increases in numbers under macroalgal cover. *C. edule*, *M. balthica*, *E.* cf. *flava* and oligochaetes showed no significant difference in abundances. Again, none of the species were significantly more abundant in the non-weed plots compared to the weed plots.

Sediments

The results of the physical properties of the sediments (% water, % organics, % silt/clay, Md ϕ and sorting coefficient) for the 4 weeks and 20 weeks samples are given in Figure 4(i) and (ii), respectively. After 4 weeks, the weed had a significant effect on all the measured sediment variables, increasing their values compared to the non-weed plots. Figure 4(ii) shows that after 20 weeks, although the weed had been slowly disappearing for nearly 2 months, the weed plots still had significantly higher values for all the sediment variables relative to the non-weed plots.

Redox potential

The redox profiles of the sediments of the weed and the non-weed plots after 4 weeks, 8 weeks and 20 weeks are shown in Figure 5(i)–(iii). After 4 weeks,

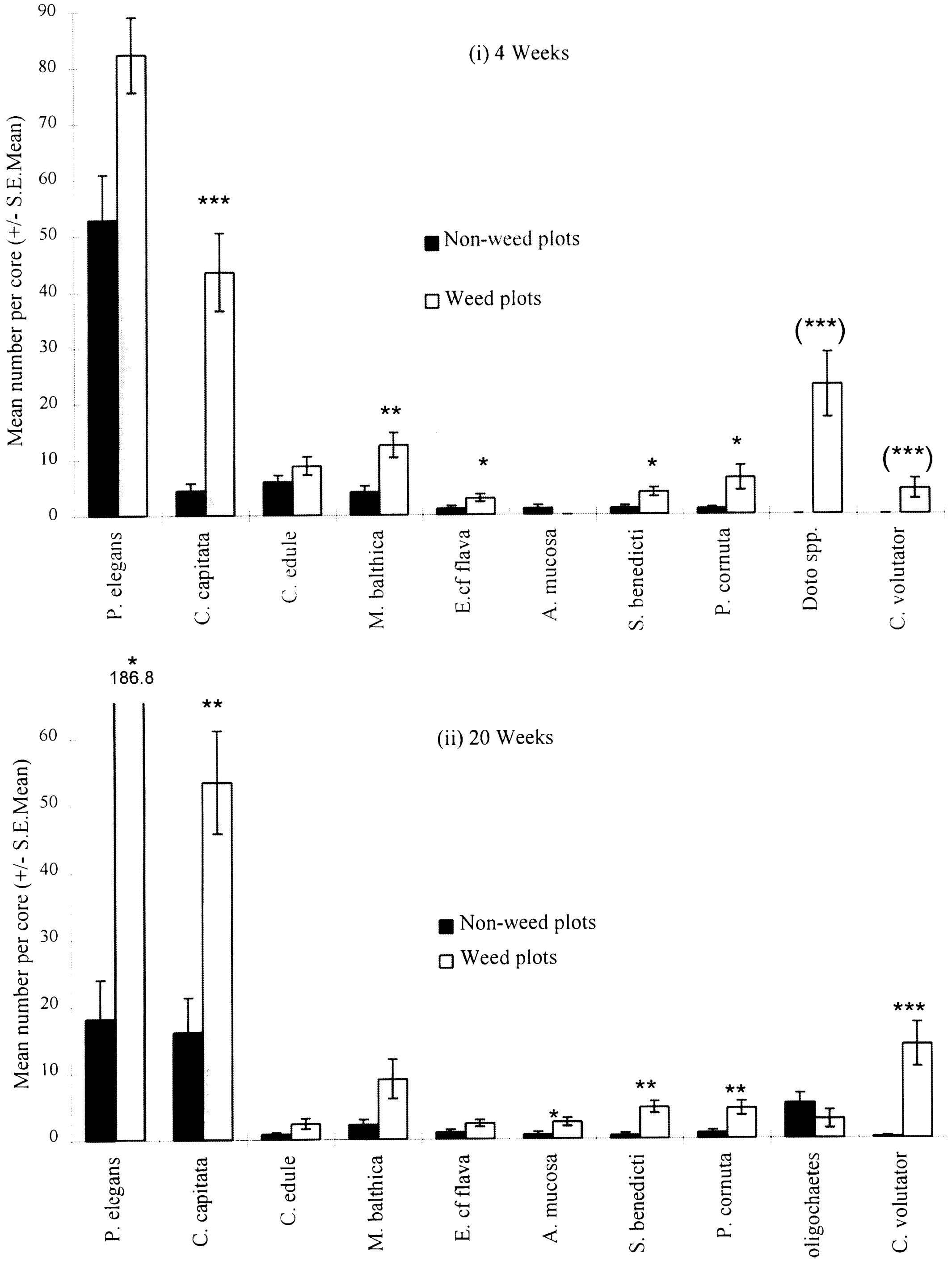

Figure 3. (i–ii) Faunal results for the weed and non-weed plots (±S.E. Mean, *n*=6); (i) 4 weeks and (ii) 20 weeks after *V. subsimplex* mat establishment. * Denotes a significant difference between the two plot types with $p<0.05$, ** denotes $p<0.01$ and *** denotes $p<0.001$, two-sample *t*-test. Brackets imply results from one-sample *t*-test.

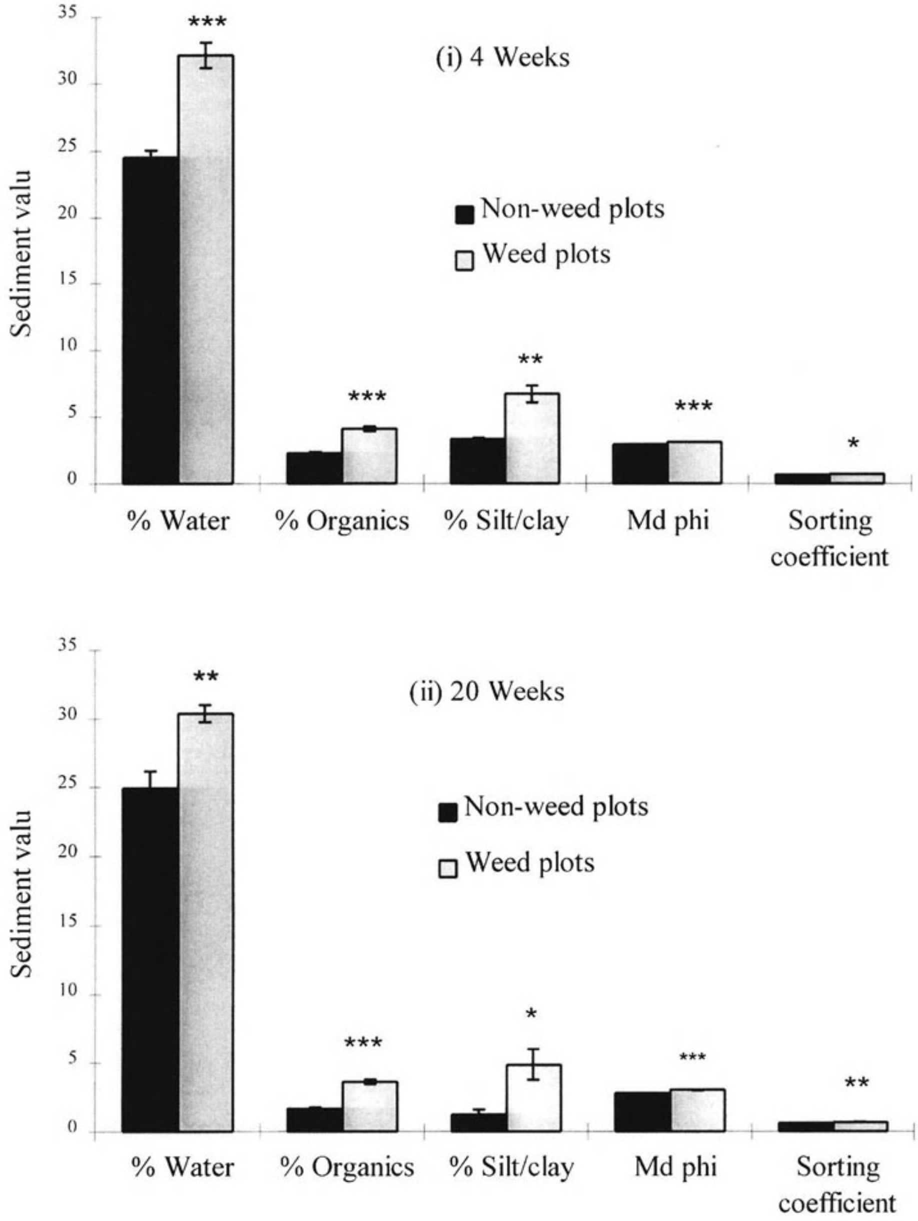

Figure 4. (i–ii) % water, organic and silt/clay contents and granulometry results for the sediments of the weed and non-weed plots (±S.E. Mean, n=6); (i) 4 weeks and (ii) 20 weeks after *V. subsimplex* mat establishment. * Denotes a significant difference between the two plot types with $p<0.05$, ** denotes $p<0.01$ and *** denotes $p<0.001$, two-sample t-test.

the sediments under the macroalgal patches became significantly more reducing compared to those of the non-weed plots. A similar response was observed for the redox profiles after 8 weeks. In January, when the weed had almost completely disappeared, the redox profiles no longer showed the significant differences shown by the physical variables at this time: the redox profiles were no longer significantly different between the two plot types (Figure 5(iii)), although the sediments of the weed plots were still slightly more reducing.

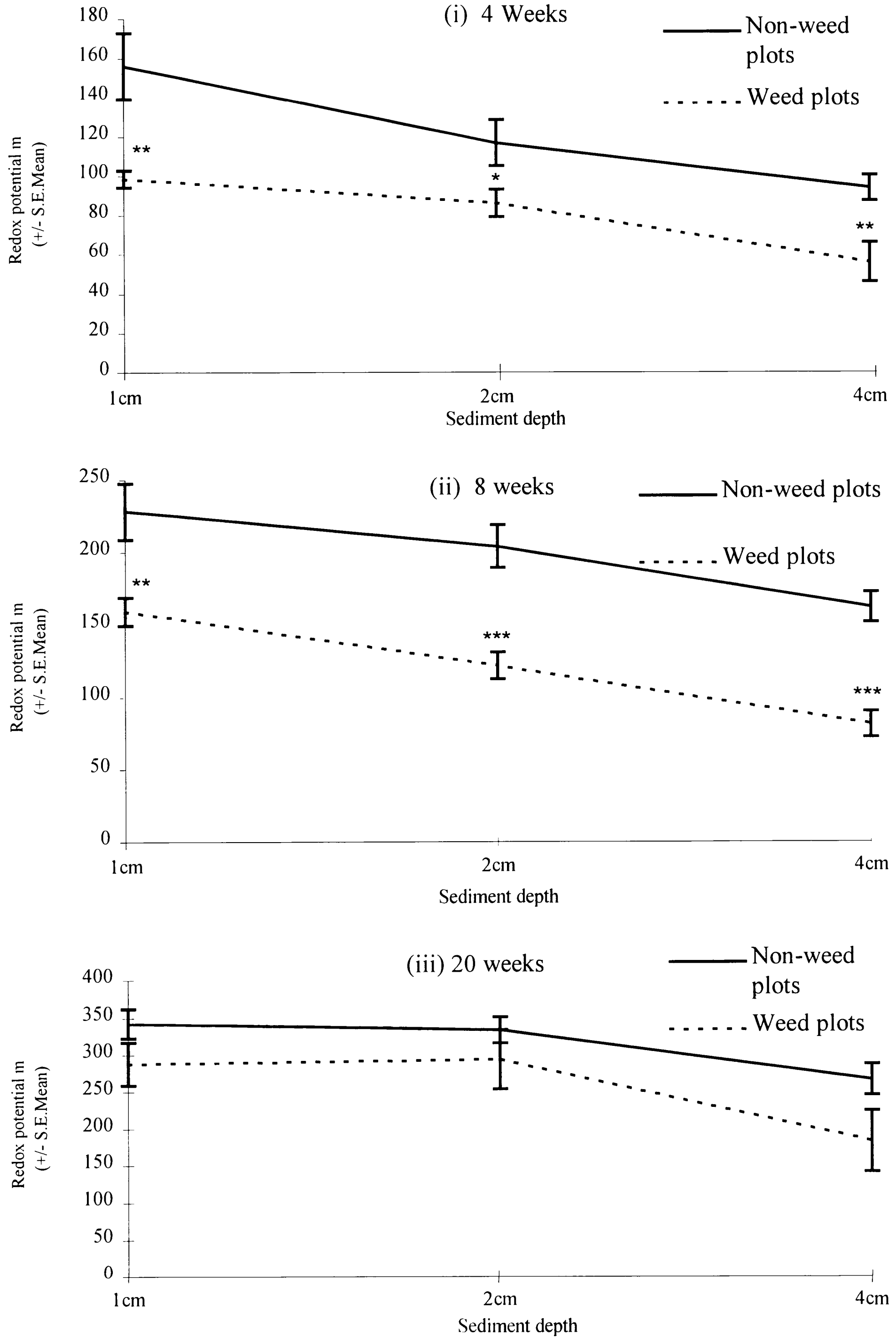

Figure 5. (i–iii) Redox potential profiles of the weed and non-weed plots (±S.E. Mean, n=6); (i) 4 weeks; (ii) 8 weeks; and (iii) 20 weeks after *V. subsimplex* mat establishment. * Denotes a significant difference between the two plot types with $p<0.05$, ** denotes $p<0.01$ and *** denotes $p<0.001$, two-sample t-test.

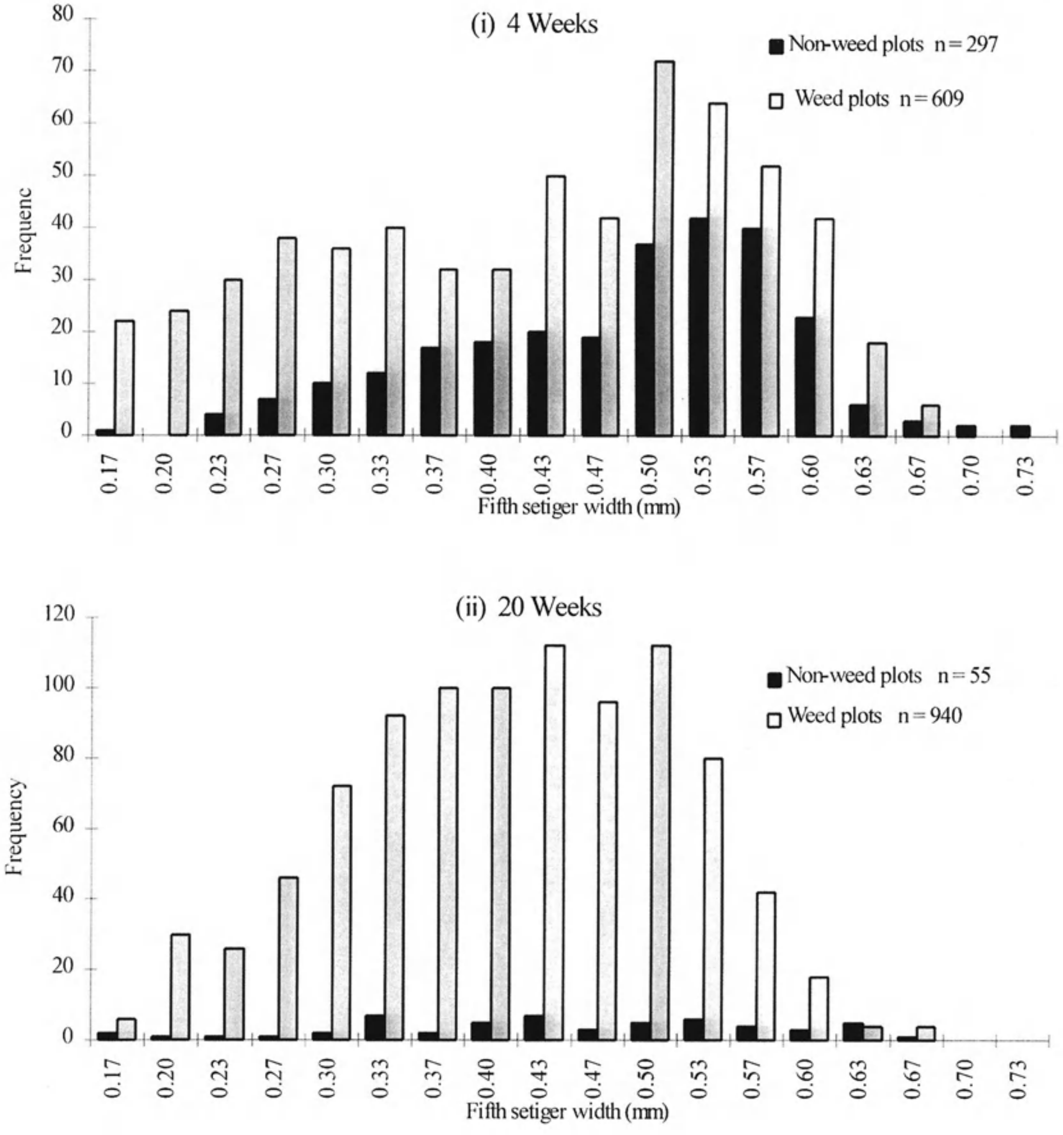

Figure 6. (i–ii) Size-frequency distributions of *P. elegans* for the weed and non-weed plots, (i) 4 weeks and (ii) 20 weeks after *V. subsimplex* mat establishment. Numbers of individuals measured are given in the legend.

Pygospio elegans *size distribution*

The 5th setiger widths of *P. elegans* for the weed and non-weed plots after 4 weeks and 20 weeks are given in Figure 6(i) and (ii), respectively. These results suggest that after 4 weeks, weed cover had a significant effect on *P. elegans* size distribution ($p<0.01$; Kolmogorov–Smirnov test) and that the main cause of this was a large increase in small individuals, 77.1% of the deviation being due to those individuals with a 5th setiger width of ≤ 0.30 mm. Therefore, the significant increase in *P. elegans* density observed after 4 weeks (Figure 3(i)) was mainly due to an increase in the number of juveniles. After 20 weeks, there was no significant difference between the size frequencies of the weed and non-weed plots ($p>0.05$; Kolmogorov–Smirnov test) although this was probably due to the low abundances in the non-weed plots preventing a definite size distribution being ascertained.

Table 2. Algal biomasses (gDW/m^2 or % cover) reported at various intertidal locations. *Indicates biomasses have been converted from fresh weight to dry weight using the conversion of Ramus & Venable (1987)

Algal species	Biomass (gDW/m^2) or % cover	Location	Authors
Enteromorpha spp.	331.6	Montrose Basin	Caudwell & Jones (1994)
Enteromorpha spp.	116.0	Eden Estuary	Owens & Stewart (1983)
Enteromorpha spp.	250.0	Lynher Estuary	Warwick et al. (1982)
Enteromorpha spp.	157.24	Ythan Estuary	Cha (unpubl.)
Enteromorpha spp. and *Ulva* spp.	42%	Langstone Harbour	Nicholls et al. (1981)
Enteromorpha spp.	21%	Firth of Clyde	Perkins & Abbott (1972)
Enteromorpha spp.	Almost complete cover	Island of Sylt	Reise (1983)
Enteromorpha spp. and *Ulva* spp.	No measure given	Medway Estuary	Wharfe (1977)
Enteromorpha spp. and *Ulva* spp.	200–400*	Langstone Harbour	Soulsby et al. (1985)
Ulva lactuca	1100.0 (1984) 300.0 (1986)	Bodega Harbor	Everett (1994)

Algal biomass

The biomass of *V. subsimplex* after 4 weeks was 49.7 gDW/m^2 and 3.4 gDW/m^2 after 20 weeks. There was never any weed present in the non-weed plots. The weed biomass observed in this study was very small compared with those reported establishing at other intertidal areas (Table 2), although many studies only give percentage cover, which is difficult to compare with quantitative biomass measurements. Furthermore, this is the first detailed investigation into the effects of the genus *Vaucheria* on the faunal invertebrates of intertidal sediments, other studies have been restricted to *Enteromorpha* and *Ulva*.

Discussion

Raffaelli et al. (1999) proposed that there was little evidence that the observed differences of macroalgal mats on infaunal species abundances were due to differences in algal morphologies, although Everett (1994) stated that this factor brought about the observed differences in faunal responses between studies. However, this controversy mainly revolves around the results of studies investigating the effects of macroalgal genera with very different morphologies from *V. subsimplex* (for example, *Enteromorpha*, *Ulva* and *Cladophora*), and therefore, studying the ecological effects of *V. subsimplex* provides important information as to the effect of macroalgal morphology in algal–faunal interactions.

The physico-chemical effects of *V. subsimplex*, which established on Drum Sands during 1997, on the underlying sediments were similar to those reported for other macroalgal mat species. These include a decreased sediment redox potential, increased silt/clay and organic contents and an increased sorting coefficient due to a reduction in water current velocity. *V. subsimplex* is a small green filamentous algae which does not possess the long filaments of other green macroalgal mat-forming genera, and therefore, has a much closer association with the sediment. Consequently, the alga formed a velvet-like carpet over the sediment (pers. obs.). If the faunal changes due to macroalgal establishment were indirect, manifested through changes to sediment conditions, one would expect that the faunal effects of *V. subsimplex* on Drum Sands would be similar to those observed in other studies. However, this was clearly not the case. While the communities resulting from the establishment of other algal taxa such as *Enteromorpha* and *Cladophora* tend to be species poor and dominated by opportunists such as *C. capitata* and oligochaetes, and the epibenthic gastropod *Hydrobia ulvae* (Pennant) (Nicholls et al., 1981; Soulsby et al., 1982), those associated with *V. subsimplex* patches were enriched, with significant increases in the abundances of the majority of species. Throughout the period of *V. subsimplex* cover, the change in actual species composition was minor – the species present in the two plot types remained similar, i.e. the increase in total abundances and diversity were mainly due to an increased abundance of those species already present.

The amphipod *C. volutator* and the nudibranch *Doto* sp. were the only taxa which were found exclusively in the weed plots while no taxa were only present in the control plots. Consequently, since the faunal responses here were markedly different from those previously reported, and the physico-chemical changes within the sediments were similar, the results of the present study suggest that macro-algae affect faunal communities directly, by their physical presence, and hence, algal morphology is likely to be a very important factor in algal–faunal interactions. This proposition is supported by the fact that *Enteromorpha prolifera* implanted onto to same area, although having very similar effects on sediment characteristics (Bolam et al., 2000), resulted in an impoverished macrofaunal community dominated by *C. capitata*.

Some of the species which increased in numbers with weed cover in this study, e.g. *C. capitata* and oligochaetes, have been found to increase under both experimentally implanted and naturally occurring weed mats (Wharfe, 1977; Nicholls et al., 1981; Wiltse et al., 1981). These opportunists increase in numbers due to the increased food supply and are relatively tolerant of reduced sediments. However, many species which have been shown to decrease in abundance under weed cover; spionids, bivalves and the amphipod *C. volutator*, for example, increased in numbers in the weed plots in this study. For example, it has been suggested that *C. volutator* suffers from the reduced sediments and/or interference with the amphipods feeding mechanism (Raffaelli et al., 1991). However, Hull (1988) found that *C. volutator* increased under experimentally implanted low *Enteromorpha* biomass and in experimental conditions where organic content was increased (Hull, 1988; Raffaelli, 2000). On the basis of his results, Hull (1988) suggested that the effect of macroalgal mats on *C. volutator* is mostly physical, the long algal filaments interfering with the normal deposit-feeding behaviour of *C. volutator* at the sediment–water interface. In contrast, as described above, the effects on *C. capitata* are probably due to an enrichment of the sediment which persisted even when the surface macroalgal material was removed. The presence of *C. volutator* in the weed plots only in our study presumably resulted from the increased food supply and the more stable sediments afforded by the weed, together with the fact that *V. subsimplex*, at least at this biomass, seems to have no deleterious physical effect on the feeding mechanisms of many deposit-feeding invertebrates. Spionids and bivalve molluscs have been shown to decline in abundance with weed cover in many studies (e.g. Perkins & Abbott, 1972; Nicholls et al., 1981; Wiltse et al., 1981; Reise, 1983; Bolam et al., 2000). Soulsby et al. (1982) found that the responses of *C. edule* and *P. elegans* were algal-biomass dependant, both species increasing with low weed biomass and decreasing with high biomass. In our study the nudibranch *Doto* sp. was found only in the weed plots and in moderate abundance. Preston (1970) proposed that nudibranchs utilise *Vaucheria* as a food source.

From our results, it is not possible to distinguish between the direct physical effects of the alga and the indirect effects through changes in sediment properties. It is clear that the biomass of *V. subsimplex* was lower than most of the quantified biomasses reported in the literature (Table 2). However, since the morphology of *V. subsimplex* is such that the small, thin filaments are found within the sediment in such a way that the alga becomes part of the sediment matrix, thus stabilising it, it is suggested that algal morphology is important when assessing effects of macro-algae on invertebrate communities. Biomass and morphology are two important variables whose specific effects cannot be separated in the present study. However, as described above, despite *V. subsimplex*'s low biomass, its overall physico-chemical effects on the underlying sediment were similar to those reported for other macroagal studies. Since the effects of *V. subsimplex* on the macrofaunal community were quite different from the ones reported in the literature for other macroagal studies, we suggest that there is a clear indication that this distinction is due to differences in algal morphology. We suggest that future studies should investigate further the role of algal morphology in algal-faunal interactions and perhaps attempt to quantify volume/biomass or volume/surface area as means to describe further algae morphology.

This study did not set out to explicitly determine the mechanisms by which many of the species increased in abundance with weed cover. Size-frequency measurements however suggested that for *P. elegans*, increased larval settlement, possibly facilitated by reduced water velocity, and/or increased juvenile survivorship, possibly due to fewer epibenthic predators, may have led to the increased densities obtained. Hull (1987, 1988) postulated that larval settlement could have accounted for the observed increases in the numbers of several species during his weed-implantation experiment. During periods of high larval availability, *P. elegans* is capable of rapidly numerically dominating after small-scale disturbances on Drum Sands and, given its planktotrophic larval reproductive mechanism, this species would probably be able to show this response to larger-scale disturbances (Bolam, 1999). The positive effect of *V. subsimplex* on *P. elegans* recruitment in this study is in contrast with the findings of the weed implantation experiment (Bolam et al., 2000) in which the main *P. elegans* larval recruitment did not successfully occur in *Enteromorpha*-implanted plots. Bolam (1999) concluded that larval filtering and the prevention of successful recolonisation by the physical disturbance caused by *E. prolifera* were the probable causes of the negative impact on *P. elegans* recruitment on Drum Sands. Perkins & Abbott (1972), Nicholls et al. (1981) and Soulsby et al. (1982) suggested that algal mats particularly caused juvenile the mortalities of various species during their surveys. It is possible that increased larval settlement and/or survival were the process(es) by which many of the other species also increased in abundance, although without size measurements and information on their reproductive strategies and population structure, this can not be concluded with any real certainty.

Conclusion

It is well documented that there are many factors which influence the effect of macroalgal cover on macrofaunal communities, the results of the present study suggest that macroalgal morphology is a major factor affecting this algal–invertebrate interaction. The results indicate that macro-algae primarily affect the fauna directly rather than via changes to sediment properties. The results also suggest that the effect of algal morphology on the macrofauna is mainly on the settling larvae: while *V. subsimplex* enhances colonisation by increasing the food supply and reducing the near-surface water velocity, other macroalgal taxa either filter-out larvae, physically disturb the sediment preventing successful colonisation or interfere with invertebrate feeding mechanisms. Future studies investigating the effects of macroalga cover on macrofaunal communities need to examine species' size distributions in order to determine causal mechanisms and whether the results obtained here are general.

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Importance of discards of a beam trawl fishery as input of organic matter into nursery areas within the Tagus estuary

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Abstract

The brown-shrimp beam trawl fishery carried out within the Tagus estuary produces discards due to the little commercial interest of most of the species caught. Between 1994 and 1996, monthly samples were collected in the two major fishing areas within the Tagus estuary, using a beam trawl, in order to estimate the amount of fish and crustaceans caught per unit of effort. The fishing effort of the commercial fleet was determined based on surveys of professional fishermen. Mortality estimates of discards were also evaluated experimentally. The main fish and crustacean species discarded after capture were *Crangon crangon* (Linnaeus, 1758), *Liza ramada* (Risso, 1826), *Carcinus maenas* (Linnaues, 1758) and *Pomatoschistus minutus* (Pallas, 1770). The estimated total amount of fishery discards in the upper part of the Tagus estuary is approximately 1500 tonnes per year, which represents ca. 90% of the captures. The mortality rate of the fishes and crustaceans discarded varied according to species and season, with the highest rates during Summer months. Considering nitrogen and carbon content of the main discarded species, an input of particulate organic matter of more than 140 tonnes of carbon and 35 tonnes of nitrogen per year were estimated for these estuarine areas.

Introduction

The beam trawl is an illegal fishing gear in Portuguese estuaries, except in the Tagus estuary where it is the most used fishing method in the uppermost areas. Despite its social and economic importance in the Tagus area, information about this fishery is very scarce (there are no catches and fishing effort estimates) and a resource assessment and management framework was never implemented.

Traditionally the beam trawl fishery catches mainly brown shrimp, *Crangon crangon* (Linnaeus, 1758). Over time, fishes such as *Solea solea* (Linnaeus, 1758) and *Solea senegalensis* Kaup, 1858, have become the main target species of this fishery, due to a decrease in the commercial value of brown shrimp. The main fishing grounds are within the nursery areas for juveniles of these fish species (Cabral & Costa, 1999).

Besides the disturbance in the sediment surface induced during hauls, the beam trawl fishery may also have a considerable impact on benthic communities (Prena et al., 1999). This fishery is characterized by a substantial bycatch of various species of fish and invertebrates (van Beek et al., 1990; Kaiser & Spencer, 1995), which are discarded immediately after sorting on board the vessels.

Several factors contribute to mortality of discarded animals, namely haul duration, conditions on deck, size of individuals and temperature (Kelle, 1976; van Beek et al., 1990). Discards may induce some changes in both benthic habitat and communities, namely an increase in the levels of particulate and dissolved organic matter which may attract scavengers and decomposers (e.g. McLusky, 1989; Prena et al., 1999).

The present work aims to estimate the weight of discards from the beam trawl fishery in the nursery areas of the Tagus estuary and the mortality rates according to species, and to evaluate its potential as input of organic matter in these estuarine areas.

Table 1. Number of fishing vessels and fishing effort (h d^{-1} $vessel^{-1}$) in the areas A and B, within the Tagus estuary

	A	B	Total
No. vessels	25	15	40
Fishing effort	4.01 (sd – 0.88)	1.48 (sd – 0.22)	3.50 (sd – 0.72)

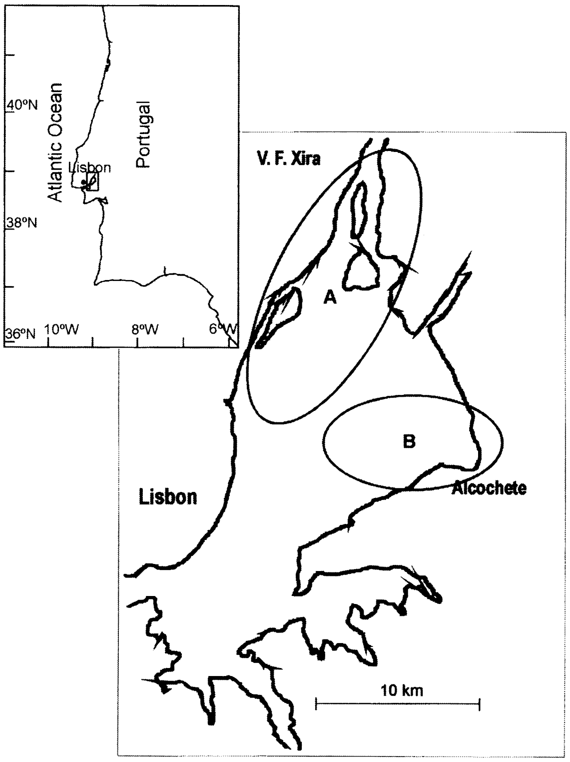

Figure 1. Location of the main nursery areas within the Tagus estuary.

Materials and methods

Study area

The Tagus estuary, with an area of 325 km², is a partially mixed estuary with a tidal range of 4.6 m. About 40% of the estuarine area is intertidal. The upper part of the estuary is shallow (less than 10 m deep) and is bordered by saltmarshes. The two main nursery areas for fish (A – Vila Franca de Xira, B – Alcochete) identified by Cabral & Costa (1999) are located in the upper estuary (Fig. 1). Although most of the environmental factors vary widely, their ranges are similar in these two areas. However, the uppermost area (A) is deeper (mean value 4.4 m) with a lower salinity (mean value 5‰) and a higher proportion of fine sand in the sediment (approximately 40%). In the other area (B), the mean values of depth and salinity are 1.9 m and 20.7‰, respectively, and the sediment is mainly composed of mud (mean value 60.4%) (Cabral & Costa 1999).

Sampling procedures and data analysis

To estimate the total weight of discards per year within the study area (Fig. 1), the fishing effort and the fish and crustacean catches had to be calculated. The fishing effort (h.day) was obtained from interviews of local fishermen. To estimate mean fish and crustacean catches per year, monthly samples were obtained during fishery independent surveys conducted in the same area during 1995 and 1996 (Cabral, 1998). Fishing effort and catch data were averaged during semi-annual periods, i.e. Spring–Summer (from April to September) and Autumn–Winter (from October to March). The proportions of the weight of discards in relation to the total weight of the catches in each of these periods were comparatively analysed using the Spearman rank correlation test, at a significance level of 0.05.

Since the calculation of total weight of discards is a function of several variables the estimation of variance is difficult. Thus, bootstrap methods were used to calculate variance estimates (Efron & Tibshirani, 1993). These estimates were calculated from 1000 bootstrap replicates obtained by random sampling with replacement.

Experiments to estimate survival of beam trawl discards were carried out on board commercial vessels operating under normal conditions in two periods (January–February 2000 and May–July 2000). After capture, discards were sorted from the catch and placed in four plastic tanks (60 × 40 × 50 cm) filled with water. Small fish and crustacean specimens were kept in two tanks (20–50 individuals in each) while large fish specimens (e.g. mullets) and crabs were placed in the other two tanks (the number of specimens per tank varied according to specimens size). The tanks were checked every 5 min and the dead specimens were recorded and removed. The experiments were terminated after 30 min, when the remaining dead and live specimens were removed. At the laboratory, the dead and live specimens of the discards at the end of each experiment, were identified, counted, measured (with 1 mm precision) and weighed (wet weight with 0.001 g precision).

Mortality rates were determined for each species. The values obtained for each one period (January–February and May–July) were compared using the Mann–Whitney test at a significance level of 0.05.

The mean mortality rates for each species and period were used to estimate the total weight of the dead fraction of discards. The variance estimate was obtained as described above.

A EUROVECTOR Elemental Analyzer was used to measure the nitrogen and carbon content of the major discarded species. The total nitrogen and carbon input in nursery areas within the Tagus estuary were estimated according to:

$$\mathrm{N_{input}} = \sum_{i=1}^{k} \sum_{p=1}^{2} \mathrm{WD}_{ip} \cdot \mathrm{MR}_{ip} \cdot \mathrm{NC}_{i}$$

and:

$$\mathrm{C_{input}} = \sum_{i=1}^{k} \sum_{p=1}^{2} \mathrm{WD}_{ip} \cdot \mathrm{MR}_{ip} \cdot \mathrm{CC}_{i}$$

respectively, where $\mathrm{N_{input}}$ is the estimate of nitrogen input, $\mathrm{C_{input}}$ is the estimate of carbon input, WD_{ip} is the weight of discards of species i in period p, MR_{ip} is the mortality rate of species i in period p, NC_i is the nitrogen content of individuals of species i and CC_i is the carbon content of individuals of species i.

Results

In the Tagus estuary, the beam trawl fishery is concentrated in the upper part of the estuary. Thirty five fishing vessels operate regularly in this area, with higher fishing effort in the northern part of the estuary (Table 1).

C. crangon, *Liza ramada* (Risso, 1826), *Carcinus maenas* (Linnaeus, 1758) and *Pomatoschistus minutus* (Pallas, 1770) were discarded at the highest rate. The values estimated for *Pomatoschistus microps* (Krøyer, 1838), *Palaemon longirostris* (H. Milne-Edwards, 1837), *Mugil cephalus* Linnaeus, 1758, *S. senegalensis*, *Dicentrarchus labrax* (Linnaeus, 1758) and *S. solea* varied between 64.9 tonnes and 12.1 tonnes per year (Table 2). *Engraulis encrasicolus* (Linnaeus, 1758), *Palaemon serratus* (Pennant, 1777), *Sardina pilchardus* (Walbaum, 1792) and *Gobius niger* (Linnaeus, 1758) presented the lowest values (less than 8.0 tonnes per year) (Table 2).

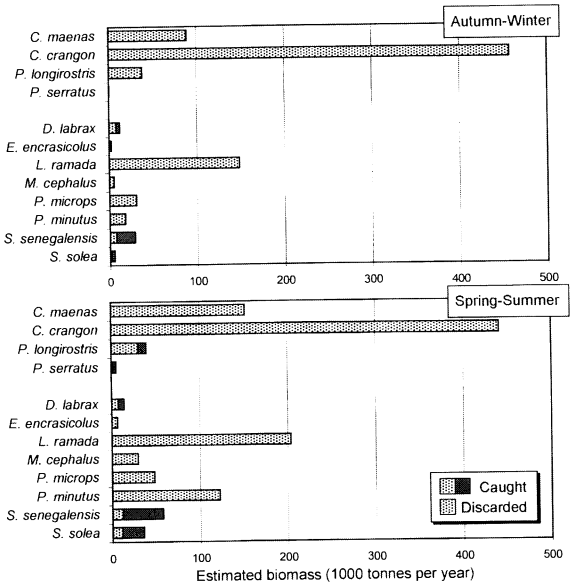

Figure 2. Estimates of biomass caught and discarded per species and period.

For the majority of the species, the proportion of discarded/caught biomass was extremely high. Only for *D. labrax*, *P. serratus*, *S. senegalensis* and *S. solea* the percent of discards represented ca. or less than 50% (Table 2).

The mean weight of discards per year, estimated based on the sampling surveys conducted in 1995 and 1996, was 1527.4 tonnes per year. This represents nearly 90% of the total annual catch (Table 2). The standard deviation of this estimate was high, reflecting considerable interannual variability.

Although the catches varied according to the period considered (Autumn–Winter and Spring–Summer), the proportion of discarded/caught biomass for each species was similar in both periods (r_S=0.96, p<0.05) (Fig. 2).

The mortality rates of the discarded organisms were generally much lower in Winter (January–February) comparatively to Spring and Summer (May–July) (Table 3). For some species, namely *C. maenas*, *S. senegalensis* and *S. solea*, the mortality rate estimates were extremely low in both periods. For the majority of the other species, the mortality

Table 2. Estimated weight of the main fish and crustaceans species caught per year and percent discarded, in area A and B of the Tagus estuary, (standard deviation between brackets)

Species	A		B		Total	
	Caught (tonnes)	Discarded (%)	Caught (tonnes)	Discarded (%)	Caught (tonnes)	Discarded (tonnes)
C. crangon	464.00	91.8	316.43	92.0	780.43	716.92
	(237.56)	(3.2)	(43.14)	(3.5)	(240.31)	(223.99)
C. maenas	109.81	95.2	102.72	92.2	212.53	199.22
	(99.79)	(4.5)	(41.60)	(6.0)	(106.99)	(101.86)
D. labrax	21.33	50.6	4.58	48.7	25.91	13.03
	(10.08)	(5.8)	(4.56)	(7.2)	(11.28)	(5.94)
E. encrasicolus	6.51	97.7	0.98	96.3	7.49	7.31
	(3.44)	(2.7)	(0.59)	(4.1)	(3.49)	(3.51)
G. niger	0.16	100.0	0.20	100.0	0.36	0.36
	(0.15)	(0.0)	(0.22)	(0.0)	(0.27)	(0.27)
L. ramada	250.47	98.7	41.11	96.8	291.59	287.07
	(62.24)	(1.7)	(27.37)	(4.5)	(72.20)	(69.65)
M. cephalus	28.17	98.7	2.80	98.5	30.97	30.57
	(25.66)	(1.7)	(1.72)	(1.7)	(25.85)	(25.45)
P. longirostris	73.64	69.8	4.46	73.4	78.10	54.69
	(27.56)	(4.9)	(1.19)	(6.3)	(27.75)	(19.93)
P. microps	62.05	100.0	2.82	100.0	64.87	64.87
	(25.05)	(0.0)	(0.43)	(0.0)	(25.33)	(24.69)
P. minutus	120.65	100.0	1.69	100.0	122.35	122.35
	(109.60)	(0.0)	(0.39)	(0.0)	(108.17)	(108.17)
P. serratus	1.28	32.5	4.55	31.4	5.83	1.84
	(1.30)	(4.4)	(3.21)	(6.6)	(5.10)	(1.60)
S. pilchardus	0.82	95.2	0.03	96.1	0.85	0.81
	(0.36)	(4.5)	(0.12)	(4.0)	(0.37)	(0.35)
S. senegalensis	62.08	18.3	24.35	19.9	86.43	16.21
	(46.41)	(6.0)	(3.93)	(4.7)	(45.91)	(9.68)
S. solea	40.42	29.9	0.36	28.7	40.78	12.15
	(15.38)	(4.5)	(0.22)	(4.9)	(15.06)	(4.99)
Total	1241.40		507.07		1748.46	1527.38
	(290.82)		(70.43)		(306.17)	(259.17)

rates determined in Winter months were low, except for *C. crangon* and *P. minutus* and *P. microps*. In the Spring–Summer period, the highest mortality rate estimate was obtained for *E. encrasicolus* (75.0%). The Summer time mortality estimates determined for *C. crangon*, *P. microps*, *P. serratus*, *P. minutus*, *P. longirostris* and *D. labrax* were also high, with values from 25.0% to 41.5%.

Considering the nitrogen and carbon contents of the most important discarded species, the beam trawl fishery is responsible for an input of ca. 140 tonnes of carbon and 35 tonnes of nitrogen per year as particulate organic matter in the nursery areas of the Tagus estuary (Table 4).

Discussion

High quantities of discards resulting from beam trawl fisheries have been pointed out by several authors (e.g. van Beek et al., 1990; Kaiser & Spencer, 1995). Fonds (1994) estimated a total of 390 thousand tonnes of dead fish and invertebrate discards in 1992 for the sole fishery in the southern North Sea. This value represents between 86% and 90% of the total annual catch

Table 4. Composition of C and N of the main fish and crustacean species and total loadings of C and N of the dead fraction of discards (standard deviation between brackets)

Species	Carbon (%)	Nitrogen (%)	Carbon (10^6 g y^{-1})	Nitrogen (10^6 g y^{-1})
C. crangon	39.18	10.10	95.74	24.67
	(1.13)	(0.22)	(19.51)	(7.42)
L. ramada	40.50	10.01	10.83	2.67
	(0.62)	(0.15)	(1.95)	(0.57)
P. longirostris	40.80	10.98	6.46	1.74
	(0.47)	(0.23)	(1.67)	(0.28)
P. minutus	40.78	11.09	17.73	4.82
	(1.19)	(0.29)	(3.41)	(0.39)
P. microps	39.15	10.38	9.19	2.58
	(1.08)	(0.26)	(2.01)	(0.43)
		Total	139.95	36.41

Table 3. Mortality rate (%) estimates determined for the most abundant species in the Autumn–Winter and Spring–Summer periods (sample size and standard deviation between brackets, respectively; * species for which differences in the two periods were significative according to the Mann–Whitney test)

Species	Autumn–Winter	Spring–Summer
*C. crangon**	5.2	41.5
	(321; 13.9)	(414; 36.1)
C. maenas	0.0	0.0
	(220; 0.0)	(198; 0.0)
*D. labrax**	0.0	25.0
	(28; 0.0)	(47; 50.0)
*E. encrasicolus**	0.0	75.0
	(31; 0.0)	(57; 50.0)
*L. ramada**	0.1	10.2
	(53; 0.2)	(44; 29.1)
*P. longirostris**	1.1	28.9
	(241; 3.3)	(182; 38.1)
*P. microps**	5.2	31.2
	(153; 8.5)	(149; 27.8)
*P. minutus**	6.7	29.2
	(114; 13.9)	(178; 41.5)
*P. serratus**	0.1	30.4
	(28; 0.2)	(49; 15.8)
S. senegalensis	0.0	0.1
	(25; 0.0)	(53; 5.0)
S. solea	0.0	0.1
	(17; 0.0)	(54; 3.2)

of target species (Fonds, 1994), which is the highest discarded rate of all fishing fleets in the North Sea (Garthe et al., 1996).

The assessment of overall discarded rate is difficult and subject to different sources of variability and errors, primarily those related to fisheries catches, landings and fishing effort. Therefore, the quantity and quality of available data determine the precision of estimates in large extent. The values determined in the present study are rough estimates that could be improved by the development of specific sampling protocols (e.g. Tamsett et al., 1999; Tamsett & Janacek, 1999).

In the Tagus estuary, the discard rates of fish and crustaceans in the beam trawl fishery were similar. In the North Sea, fish by-catch can be considerable, particularly for some flatfish species, e.g. *Limanda limanda*, *Pleuronectes platessa*, *Platichthys flesus* and *S. solea* (Robin, 1992; Garthe et al., 1996; Berghahn & Purps, 1998).

Survival of discards differs greatly according to species. Kaiser & Spencer (1995) concluded that molluscs and epibenthic crustaceans were highly resistant to the effects of capture, while the majority of the fish species and swimming crabs suffered high mortalities. Several studies on discards survival outlined that mortality of small fish was quite high (Fonds et al., 1992; Kaiser & Spencer, 1995). This was also found in the experiments carried out in the present study, except for *S. solea* and *S. senegalensis*.

As previously reported by Kelle (1976) and van Beek et al. (1990), temperature is extremely important to discard mortality. All the mortality estimates determined in the experiments conducted in the Spring–Summer period were higher than those obtained in Autumn–Winter.

Beam trawl fisheries have a significant impact in benthic communities and cause damage to both epifauna and infauna (e.g. Bergman & Hup, 1992). Furthermore, due to the low selectivity of this gear, juveniles are heavily caught, which could contribute to a decrease in the stocks of several marine fishes. However, several authors (e.g. Kennelly, 1995) suggest that mortality of juveniles discarded by beam trawlers is low when compared with natural mortality. According to our results, the impact of the beam trawl fishery on recruitment may vary considerably according to species.

The dead organisms left in estuarine grounds due to discarding may induce several changes in both abiotic and biotic characteristics of benthic habitats. Caddy (1973), Kaiser & Spencer (1994) and Prena et al. (1999) reported that scavengers are attracted to these disturbed areas, where they benefit from the increased availability of food.

Several studies demonstrated the importance to seabirds of fishery waste (e.g. Blaber et al., 1995; Walter & Becker, 1997). Garthe et al. (1996) found that discards represent a considerable proportion of the food of seabirds in the North Sea.

Discards also increase the amount of biogenic material in estuarine grounds, which promotes microbial decomposition. These decaying remains have a valuable role in stabilising estuarine systems by levelling out the seasonal variation in primary production and ensuring a year-round nutrient supply (McLusky, 1989). However, the pathways of organic input to estuarine environments are extremely dependent on water renewal and quality, as are primary and secondary production. It has been reported for several estuarine systems that an increase in nutrients loadings may induce eutrophication, causing algal blooms and depletion of dissolved oxygen (Howarth et al., 2000). These changes alter the dominant biogeochemical processes. In particular, an increase in nitrogen loading increases denitrification, which causes a decrease in N:P ratio and hence a nitrogen versus phosphorus limitation for primary production (Seitzinger, 2000).

The chemical composition of decaying remains changes during the decomposition process and thus there is not a direct relationship between the carbon and nitrogen content of discarded organisms and the resulting nutrients available for primary producers and animal consumption.

Measures to reduce discards are extremely important and should be promoted. The two commonest ways to achieve this that are usually pointed out are an increase in mesh size of fishing gears and a reduction of the fishing effort (Anonymous, 1994). Since the beam trawl fishery in the Tagus estuary is supposed to catch brown shrimp, an increase in mesh size would hardly be considered. A reduction of the fishing effort in some particular periods would surely reduce discards, since the amount and mortality of discards is highly dependent on season and time of day. The unique legislative status of the Tagus estuary, concerning the use of beam trawl, should also be reviewed.

Further studies should be focused on the factors that influence discard mortality and impacts on estuarine communities.

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The intertidal soft-bottom infaunal macrobenthos in three Basque estuaries (Gulf of Biscay): a feeding guild approach

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Abstract

Feeding types of the infaunal macrobenthos of the intertidal soft-bottom communities from the estuaries of La Arena, Plentzia and Gernika, along the Basque coast (Gulf of Biscay) were studied in relation to habitat conditions, characteristics of each estuary and seasonal variation. General feeding structure observed in La Arena estuary was different from the other estuaries: omnivores were dominant, both in number of species and individuals, while surface deposit-feeders were the main feeding group in Plentzia and Gernika both in the finest sediment communities and in the sandy ones. Subsurface deposit-feeders were abundant in Plentzia and Gernika but scarce in La Arena estuary. Feeding types were related to the environmental variables. Sandy sediment communities presented a more balanced proportion among different feeding types and showed higher values of feeding diversity; suspension-feeders and predators were abundant only in that communities. By estuaries, feeding diversity was higher in Gernika and lower in La Arena. Seasonal variation in feeding diversity was observed, explained by changes in density of certain feeding groups; a common pattern of variation was found in Gernika communities, with an increase in June and a decrease in September. Oscillation of this index in Plentzia and La Arena depended on the estuary and the different communities; nevertheless a decrease of feeding diversity in September was commonly observed. Finally, feeding structure observed in our estuaries was compared to those described in coastal areas with organic enriched sediments, in order to assess the degree of perturbation of the environment based on the relative dominance of a certain feeding type in the macrobenthos.

Introduction

The use of functional groups, defined by feeding type, motility type and method of food capture, has been widely extended during the last years for several purposes. Macrobenthos (polychaetes are the only group included in many of the works) is classified into categories in studies of energy flows in marine sediments (Gaston & Nasci, 1988; Monaco & Ulanowicz, 1997) and control of benthic communities (Posey et al., 1995), for the assessment of pollution impact (Maurer & Leathem, 1981; Dauer, 1984a; Hily, 1991; Carrasco & Carbajal, 1998) or in the analysis of the effects of environmental gradients on marine benthic communities (Gray et al., 1988; Bonsdorff & Pearson, 1999).

The intertidal soft-bottom endofauna of the estuaries of La Arena, Plentzia and Gernika, (Gulf of Biscay, Northern Iberian Peninsula) was studied in a previous work (García-Arberas, 1999). Differences among the three estuaries were observed in relation to abiotic and faunistic data. In general, benthic assemblages of La Arena estuary were unlike to those found in the Plentzia and Gernika estuaries, even in the same sedimentary bottoms, and La Arena could be considered as a poor estuary, of low diversity and density. In this work, feeding types of the intertidal soft-bottom communities of these three estuaries were analysed in relation to other faunistic data, habitat conditions (granulometry, organic matter content, etc.) and to the characteristics of each estuary. Seasonal variation of feeding groups and feeding diversity were also evaluated. Finally, our results were compared to those described in coastal areas with organic enriched sediments, in order to evaluate the use of this approach in the assessment of the degree of perturbation of the en-

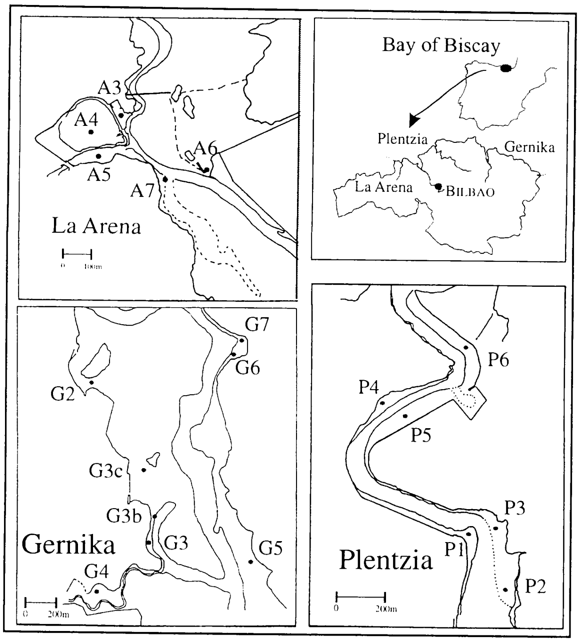

Figure 1. Location of the sampling stations in the estuaries of La Arena, Gernika and Plentzia. Situation of the three estuaries in the province of Biscay, in the North Iberian Peninsula.

vironment based on the relative dominance of a certain feeding type in the macrobenthos.

Materials and methods

The estuaries of La Arena (43° 20′ N, 3° 07′ W), Plentzia (43° 24′ N, 2° 55′ W) and Gernika (43° 22′ N, 2° 40′ W) are small and shallow estuaries located near Bilbao, in the province of Biscay in the Basque Country (Fig. 1). In the basins of the estuaries of Plentzia and Gernika some minor industrial activity takes place but most of the human activity belongs to the primary sector. This sector is also predominant in the basin of the estuary of La Arena, but a petrochemical complex is located lower down. Both the Plentzia and Gernika estuaries can be considered as basins with a slight degree of pollution while La Arena is considered as a basin with intermediate pollution (Borja et al., 1998).

A total of 74 samples were taken over February, March, June, September and December 1989 in the mid-littoral intertidal area in the lower and middle estuary: at five fixed stations in La Arena, six in Plentzia and eight in Gernika (Fig. 1). On each sampling occasion, sediment was removed at low tide from quadrants measuring 40×40 cm to a depth of 25 cm, sieved (1 mm mesh screen), and fixed in 4% formalin-sea water solution. Temperature, pH, oxygen, redox and salinity of the emerging water when digging were also measured using field standard apparatus. In the laboratory, faunal samples were sorted and individuals identified to species level when possible. Sediment analyses, using standard methods, include percentage content of sedimentary fractions: coarse elements, coarse sand and fine sand, silt and clay, porosity and humidity and organic matter content (method of Walkley & Black). Several data matrix were obtained (biotic and abiotic data) and univariate and multivariate analysis were employed. Data were transformed and normalised when required. Ordination and classification analysis and also cenotic indexes were employed in order to find out and characterise the soft-bottom communities in the intertidal zone of the three estuaries. Methodology is discussed with more details in García-Arberas (1999).

Assigning trophic status to species is subject to error, due to a lack of information on specific feeding types, higher in some faunistic groups, and to the variability of feeding types employed by individual at genus or even species level. There are species that can switch between deposit feeding and suspension feeding depending on near-bottom flow conditions as some spionids (Dauer, 1984b; Taghon, 1992; Gaston et al., 1988) and bivalves (Fallensen, 1992). We have considered five feeding types: Surface deposit-feeders (SDF), Subsurface deposit-feeders (SsDF), Predators (PRED), Suspension-feeders (SUSP) and Omnivores (OMNI). Herbivores were very scarce in the studied bottoms and they were considered, with the unknown classifications, as OTHER, and usually not included in the analyses. Species were classified following Fauchald & Jumars (1979), Whitlatch (1981), Commito & Ambrose (1985), Whitlatch & Zajac (1985), Frid & James (1988), Eleftheriou & Basford (1989), Beukema (1991), Lardicci et al. (1992) and Sánchez Mata et al. (1993).

Feeding structure (in terms of number of species and number of individuals of each feeding type), was spatially and seasonally analysed. A feeding diversity index was estimated, applying the Shannon diversity index to our data ($H' = -\Sigma p_i \log_2 p_i$; Pielou, 1966). Data on the abundance of feeding groups were analysed employing Canonical Correspondence Analysis (CCA), a multivariate statistical method of ordination, using the CANOCO program (ver. 4) (Ter Braak & Smilauer, 1998). Abundance of each feeding type was first transformed ($\mathrm{Ln}(x + 1)$). Using this technique the biological data are arranged on an environmental

Table 1. Benthic communities described in the estuaries of La Arena, Plentzia and Gernika. Sampling stations are included in brackets. Species characterise that bottoms by their fidelity or/and dominance (P=Polychaeta, M=Mollusc, C=Crustacean, I=Insect larva). Some sediment characteristics and average values of community descriptors are included (Q_{50} median grain size, OM organic matter; R= No. species/sample; D=Density, H=Shannon diversity index). Last column corresponds to average values of feeding diversity index

Communities	Characteristic species	Q_{50} (μ)	OM (%)	R No. sp	D (ind m^{-2})	H (Bits)	H feed (Bits)
La Arena							
Sandy (A6)	*P. ulvae* (M)	565	0.61	3	374	1.06	0.87
Muddy sand (A5)	*S. benedicti* (P) & oligochaeta	122	2.36	9	2328	1.89	1.35
Clayey (A3, A4, A7)	*H. diversicolor* (P) & Tipulidae (I)	54	0.55	6	603	1.57	1.07
Plentzia							
Coarse sand (P1, P6)	*S. armiger* (P) & *E. pulchra* (C)	1108	0.13	7	471	1.69	1.24
Sandy (P5)	*C. edule* (M)	488	0.53	11	772	2.12	1.54
Muddy sand (P2, P3, P4)	*P. ulvae* (M) & *H. diversicolor* (P)	75	2.09	14	2729	2.30	1.30
Gernika							
Sandy with coarse elements (G2)	*A. oxycephala* (P)	1377	0.22	17	689	2.89	1.94
Coarse sand (G3b, G3c)	*S. martinensis* (P) & *S. armiger* (P)	1014	0.12	10	393	2.50	1.80
Sandy (G6)	*E. oerstedi* (P)	407	0.89	20	1032	2.91	1.57
Muddy sand (G3, G4, G5)	*H. diversicolor* (P), Dolichopodidae (I) & *P. ulvae* (M)	146	2.15	12	3364	1.93	1.48
Clayey (G7)	*A. romijni* (P)	54	2.65	6	603	1.57	1.12

basis, offering in a single diagram the direct interpretation of possible relationships between species (in our case feeding types), stations and environmental variables (Ter Braak & Prentice, 1988; Gamito, 1997). This program allows us to evaluate the possible influence of categorical variables (DUMMY), such as the estuary and the seasonal variation. The distribution of feeding types was also compared with other benthic communities descriptors, as sample richness, density, Shannon diversity and evenness. The relationship between environmental and categorical variables was tested by the Monte-Carlo permutation test and Pearson correlation analysis was also employed.

Results

Faunistic global results obtained can be resumed as a total of 2707 individuals collected in 21 samples and belonging to 22 species in the La Arena estuary, 6258 individuals of 48 species in 23 samples analysed in the Plentzia estuary and 8785 individuals of 88 species in the 30 samples collected in Gernika. Total data matrix, faunistic results joined to environmental variables, were treated with several univariate and multivariate analyses (García-Arberas, 1999), leading us to consider 11 infaunal communities in the three estuaries, named in relation to the type of sediment. We distinguished, from finer to coarser areas, the clayey community (in La Arena and Gernika), the muddy sand and the sandy communities (in the three estuaries), the coarse sand community (in Plentzia and Gernika), and the sandy with coarse elements community (in Gernika). Some of their characteristics are resumed in Table 1, including several sediment parameters (sampling stations are indicated in Fig. 1). Distinctive species were chosen according to their dominance or/and their degree of exclusivity in the community.

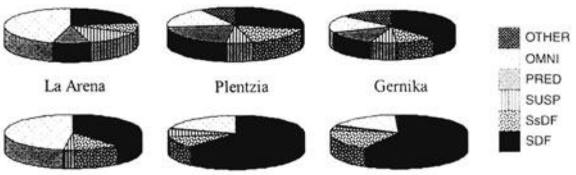

Figure 3. Feeding structure of the estuaries of La Arena, Plentzia and Gernika. Percentage of feeding types, in number of species (above) and individuals (below).

Table 2. Significant Pearson correlation coefficients performed between feeding types abundance and environmental variables, from La Arena, Plentzia and Gernika estuaries (N=74; p<0.01, *p<0.05). SDF=Surface deposit-feeders, SsDF=Subsurface deposit-feeders, SUSP=Suspension-feeders, OMNI=Omnivores, PRED=Predators

Environmental variables	Feeding types				
	SDF	SsDF	SUSP	OMNI	PRED
pH				−0.267*	0.280
Salinity (%)	−0.405	−0.384	0.249*	−0.414	0.240*
Oxygen (mg/l)	−0.547	−0.400			
Eh (mV)	−0.469	−0.353			
Coarse elements (%)				−0.306	
Coarse sand (%)	−0.320			−0.432	0.251*
Fine sand (%)	0.329			0.566	
Silt (%)	0.350		−0.236*	0.396	
Clay (%)			−0.248*	0.308	
Median grain size (μ)	−0.296			−0.419	
Porosity (%)	0.344	0.286		0.406	
Humidity (%)	0.386	0.239*		0.372	
Organic matter (%)	0.513	0.350		0.559	

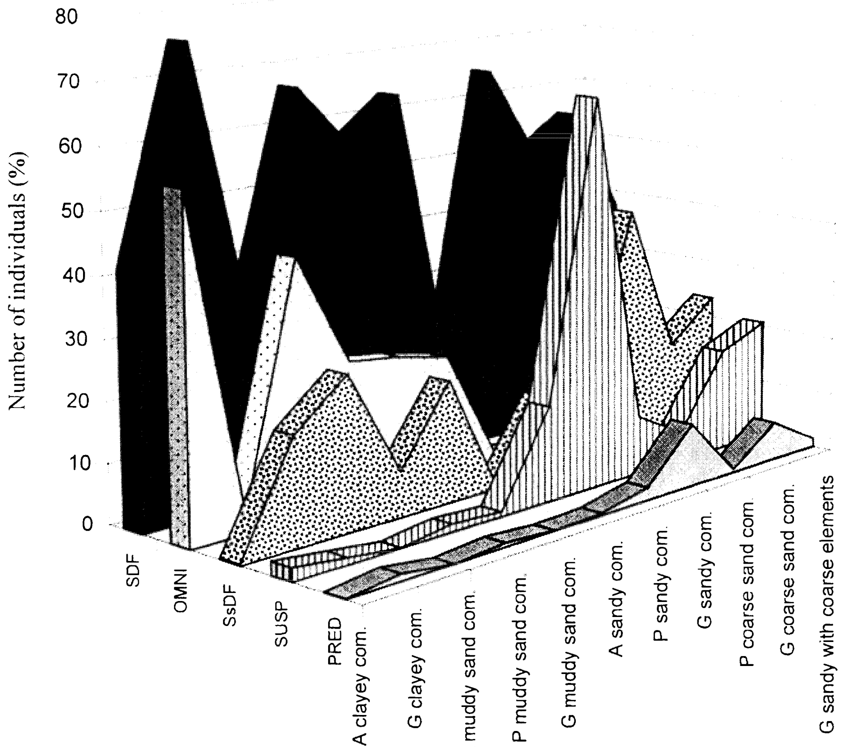

Figure 2. Percentage of density of each feeding type in the benthic communities of the three estuaries. Communities are ordered by granulometry: those from finer bottoms (left) to those from coarsest sediments (right) (A=La Arena; P=Plentzia; G=Gernika).

General feeding structure of the three estuaries was compared, analysing the number of species and individuals of each feeding type (Fig. 2). Feeding types observed in La Arena estuary were different from those of Plentzia and Gernika. Omnivores were the feeding type dominant in La Arena, both in richness (10 species) and abundance (1191 individuals), and the main faunistic groups were insects larvae and polychaetes, respectively. The number of species belonging to other feeding categories was similar, but surface deposit-feeders were the second feeding type in terms of abundance (1005 individuals). The main feeding type in Plentzia and Gernika corresponded to surface deposit-feeders, due to polychaetes in the case of number of species and molluscs in density; surface deposit-feeders were clearly dominant in abundance (3829 individuals in Plentzia and 5106 individuals in Gernika), while feeding structure was more balanced in relation to the number of species of each category. Subsurface deposit-feeders were of relative importance both in terms of richness and density, there being no great differences among estuaries; annelids were the only faunistic group which showed that type of feeding. There were species behaving as suspension-feeders and predators in the three estuaries, but usually in a small number of individuals, mainly predators.

Feeding types of the benthic communities described in each estuary were also analysed. Percentage of the number of individuals belonging to the different type of feeding is shown in Figure 3. Relationship between feeding types and environmental variables was studied by Pearson correlation analysis and significant coefficients are indicated in Table 2.

Surface deposit-feeding was the main feeding type in almost all the communities. In some cases with high values of dominance, being more than the 60% of the total density. This fact was observed not only in the fine sediment communities but also in the sandy ones, as it occurred in the clayey community from Gernika, the muddy sand community from Plentzia and the sandy communities from Gernika and La Arena. However, the abundance of this feeding type was positively correlated with fine sand and silt content of the sediment and related variables, as organic matter content (Table 2). The main surface deposit-feeders, inhabiting most of the area studied, were the mollusc *Peringia ulvae* (Pennant, 1777) and *Scrobicularia plana* (Da Costa, 1778) and the polychaete *Streblospio benedicti* (Webster, 1879). Other surface deposit-feeders showed high abundance depending on the communities, as *Polydora ciliata* (Johnston, 1838) and *Alkmaria romijni* (Horst, 1919) in the case of the clayey community from Gernika. They were mainly polychaetes, basically spionids.

Subsurface deposit-feeders were abundant in Plentzia and Gernika but scarce in La Arena estuary, excepting in the muddy sand community. They did not show a significant relationship with the sediment granulometry but were correlated with other environmental variables, as the organic matter content of the sediment (positively) or the salinity and oxygen of the running water (negatively). The main subsurface deposit-feeders were oligochaetes and polychaetes belonging to the family Capitellidae. Their dominance was higher in the coarse sand community from the Plentzia estuary (41.7%), basically due to the polychaetes *Capitella capitata* (Fabricius, 1780) and *Scoloplos armiger* (Müller, 1776).

Suspension-feeders could inhabit the fine sediment areas but their presence was considerable only in sandy communities, being the feeding type clearly dominant in the case of that of the Plentzia estuary (64.8%). Their abundance showed very few significant correlations with the environmental variables; the exception being with salinity (positively) and with the percentage of silt and clay of the sediment (negatively). The most abundant suspensivore species was the bivalve *Cerastoderma edule* (Linnaeus, 1767).

Predators were poorly represented in the three estuaries. They only inhabited some of the sandy communities, showing a certain relative abundance just in two of them, particularly in the Gernika estuary. A significant relationship (positive) between predators abundance and environmental variables was found only for the case of the coarse sand content of the sediment and the salinity and pH of the running water. The main predators were basically polychaetes, as *Nephtys hombergii* (Savigni, 1818), *Phyllodoce (Anaitides) mucosa* (Oersted, 1834) or *Glycera tridactyla* (Schmarda, 1861).

Omnivores were present in almost all the samples, showing a high variation in their relative abundance, even within the same community. This type of feeding showed a great number of significant correlations with the environmental variables (Table 2). Generally, a negative relationship with the coarse sediment and other related variables was observed, while they appeared positively correlated with the fine fractions of the sediment. Omnivores were clearly dominant in two communities from La Arena estuary: the clayey and the muddy sand communities. The main omnivores were the polychaete *Hediste diversicolor* (Müller,

1776) and the crustacean *Cyathura carinata* (Kroyer, 1847).

Feeding diversity was analysed for benthic communities of the three estuaries; averaged values are included in Table 1. Lower values were close to 1 bit and estimated for the sandy community from La Arena estuary and for the clayey communities from La Arena and Gernika. The greatest values, close to 2 bits, were obtained for two of the communities described in Gernika: the sandy with coarse elements and the coarse sand ones. This index seemed to be related to sediment characteristics, being lower in muddy environments. Concerning estuaries, the feeding diversity was higher in Gernika and lower in La Arena, even in sandy sediment communities.

Seasonal oscillations in density of feeding types were observed. They depended on communities and estuaries but, in general, surface deposit-feeders were the most variable in number of individuals. In order to find a possible seasonal pattern of variation of the feeding structure, evolution of the feeding diversity index was also analysed (Fig. 4). This index varied little in La Arena estuary, except for a decrease observed in September in the muddy sand community and in December in the sandy one. In Plentzia, feeding diversity was similar throughout the time in the muddy sand community but showed more variation in the two sandy communities, oscillations being opposite. Finally, a seasonal trend of variation was observed in the Gernika estuary, with a general increase of feeding diversity in June and a decrease in September.

Ordination diagrams obtained from the Canonical Correspondence Analyses (CCA) are represented in Figures 5 and 6, where arrows indicate the importance of each variable included in the analysis. All the analyses were found to be significant (Monte-Carlo test $p \leq 0.001$), except for seasonal variation, but a great difference was observed in the percentage of the variance that each analysis explained (Table 3). Differences in the feeding types among estuaries and seasons have been mentioned above, being also reflected in the ordination analysis (Fig. 5A,B). On the other hand, subsurface deposit-feeders seemed to be associated with samples having high values of density and also richness and diversity (Fig. 5C), while suspension and predation feeding types would be associated with the last two community descriptors. However, the environmental variables would explain the higher percentage of the variability of the feeding types abundance. In Figure 6, a granulometrical gradient is observed, with fine sand, silt and clay as opposed to coarse sand. Omnivores would be associated with finer sediments while suspensivores and predators with coarse sand content (and also temperature and salinity). These results agreed with those found by the Pearson correlation analysis. CCA showed a second gradient defined by pH and coarse elements as opposed to oxygen content. Subsurface deposit-feeders would be related to sediments with a high percentage of coarse elements but also with high levels of organic matter. In that sense, the abundance of this type of feeding was positively correlated with the organic matter content of the sediment but not with its granulometry. Surface deposit-feeders were located in the origin of the coordinates of the diagram due to the ubiquity of this feeding type in the area under study.

Discussion

Detritivores, represented by surface and subsurface deposit-feeders, are generally the dominant trophic group in infralitoral soft bottom areas of estuarine and coastal ecosystems (Gaston & Nasci, 1988; Lastra et al., 1991; Muniz & Pires, 1999). In the intertidal soft-bottoms of the estuaries of Plentzia and Gernika, surface deposit-feeding was the main feeding type in all the benthic communities, with the exception of the sandy community in Plentzia where suspensivore abundance was higher. In La Arena estuary, a different feeding structure was found, surface deposit-feeders being dominant only in the muddy sand community while omnivores were the main feeding type.

Feeding types were clearly related to the environmental variables. Physical parameters as hydrodynamics, sediment grain size or quantity and quality of the available food (Sanders, 1958; Whitlatch, 1977, 1981; Maurer & Leathem, 1981; Cocito et al., 1990), have been indicated as factors determining the dominance of feeding types. Deposit-feeders are favoured in muddy and muddy sand low-energy environments while suspension-feeders prevail in sediments with low content of fine fractions (Sanders, 1958; Whitlatch, 1977; Dewarumez, 1983). A clear example of this fact is observed by Dewarumez et al. (1992) who found a ‘transition assemblage’ between the pebbles community, dominated by suspensivores, and the *Abra alba* community, dominated by deposit-feeders; the former was composed of suspension-feeders and deposit-feeders. In our study, suspension-feeder dominance was higher in sandy areas although percentages changed a lot according to communities, the main

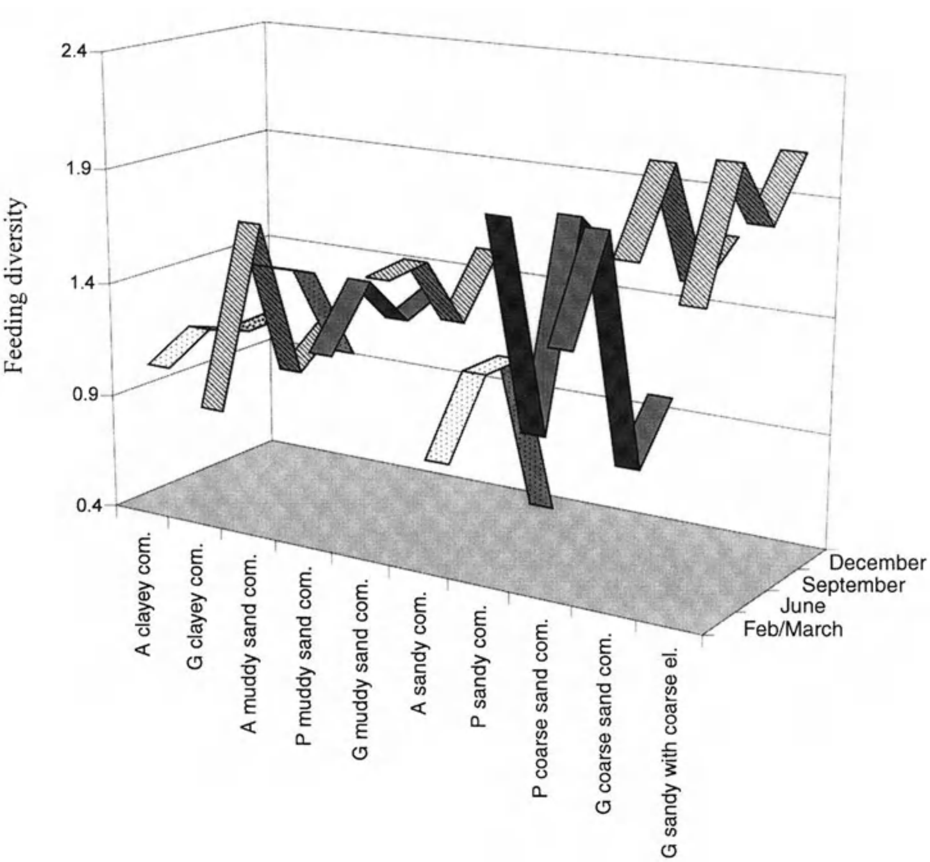

Figure 4. Seasonal oscillations of feeding diversity index, differentiating each community and estuary. Communities are ordered by granulometry: those from finer bottoms (left) to those from coarsest sediments (right) (A=La Arena, dotted; P=Plentzia, grey; G=Gernika, lined).

Table 3. Canonical Correspondence Analysis (CCA). Eigenvalues of the two first axis of each analysis and percentage of the variance explained. Significance of the first canonical axis (Monte-Carlo test, *p*-value) is included

Analysed variable	Axis	Eigenvalue	Significance	% Cumulative variance
Seasonal variation	I	0.019	0.055	5.3
	II	0.006	–	6.8
Estuary	I	0.068	0.005	18.7
	II	0.003	–	19.5
Community indexes	I	0.060	0.005	16.4
	II	0.016	–	20.8
Environmental variables	I	0.095	0.005	26.1
	II	0.037	–	36.1

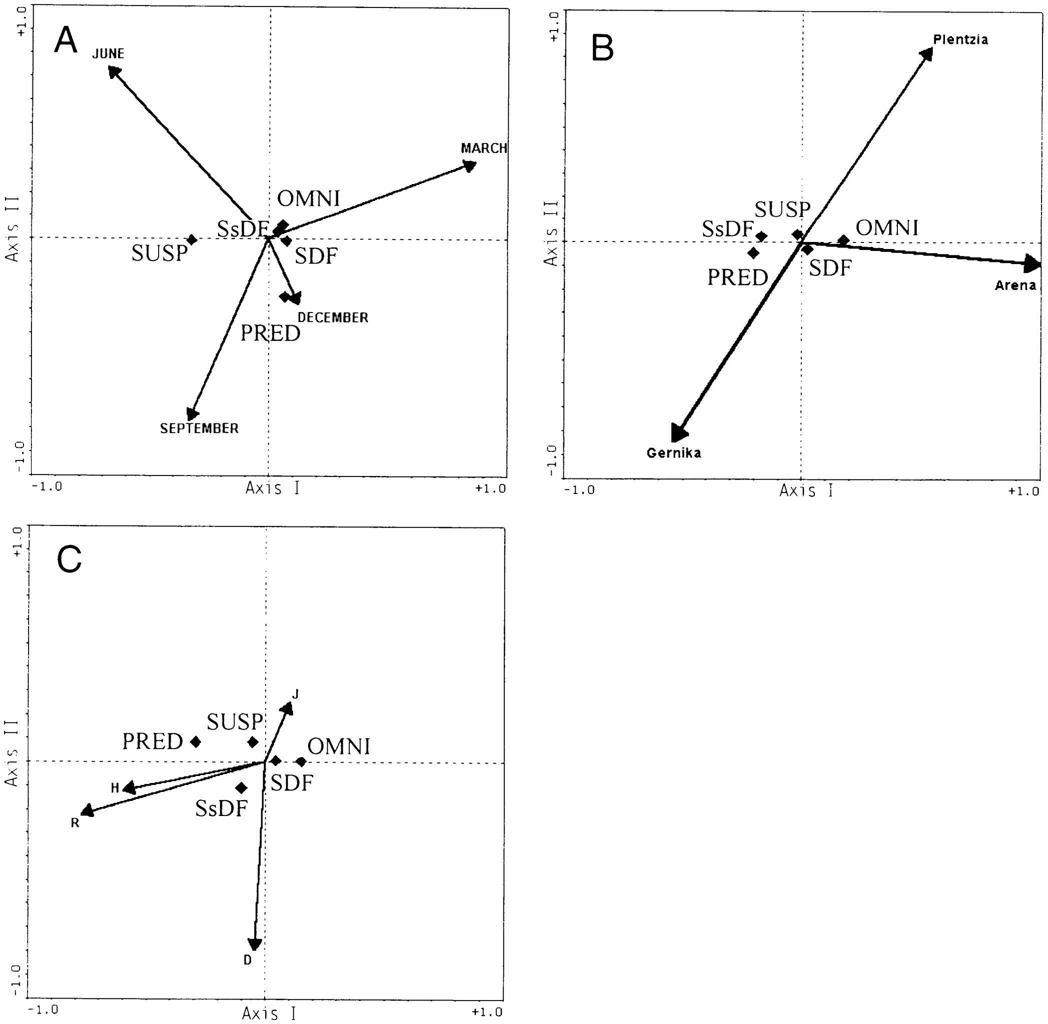

Figure 5. Ordination diagrams obtained from the Canonical Correspondence Analysis (CCA). In the figures, feeding types are located in relation to the influence of: (A) seasonal variation, (B) estuary, (C) community indexes (R=Richness, D=Density, H=Shannon Diversity, J=Evenness).

feeding type being only in one of them. They showed a significant negative relationship with the silt and clay sediment content. By contrast, surface deposit-feeder abundance was positively correlated with sediment fine fraction and related variables, such as organic matter content.

In the three estuaries, sometimes one type of feeding was clearly dominant in the community, but generally, a second or even a third feeding type showed a considerable percentage of density. Sandy bottom communities showed a more balanced proportion among different feeding types, mainly in the Gernika estuary. This involved higher values of feeding diversity. Maximal values of this index were estimated in the typically sandy communities in Gernika, where the predators percentage was also the largest. This type of feeding is more frequent in coarse sediments with low levels of mud (Maurer & Leathem, 1981; Gaston & Nasci, 1988), as confirmed by the CCA and the correlation analysis. This fact could be explained by a higher diversity of microhabitats in coarse sediments (Carrasco & Carbajal, 1998) and because the proliferation of potential prey organisms occurs in their interspaces (Muniz & Pires, 1999). Predators were also associated with samples having high values of richness and diversity.

Apart from hydrographic and sediment conditions, oscillations observed in densities of feeding types can

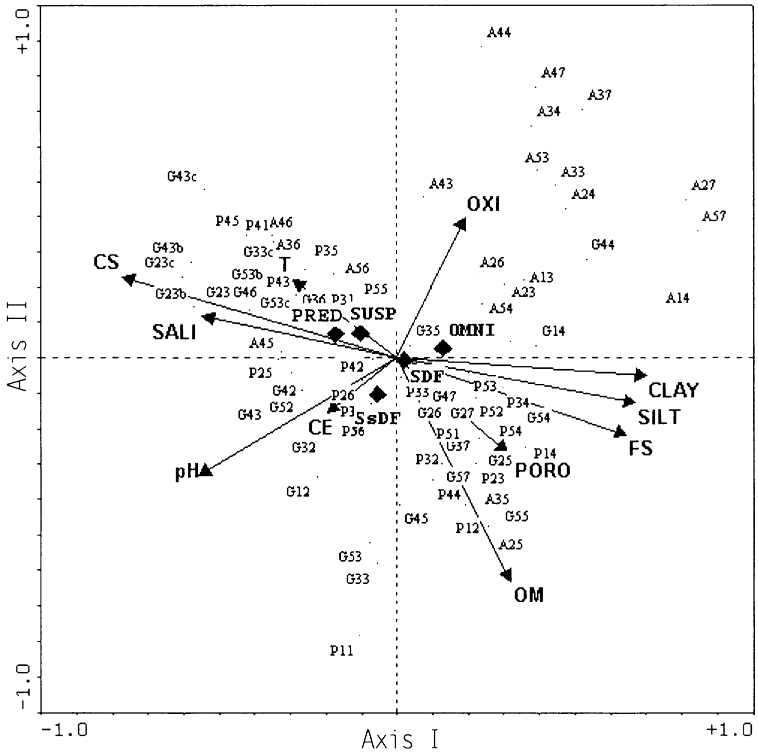

Figure 6. Ordination diagram obtained from the Canonical Correspondence Analysis (CCA). Feeding types are located in relation to the influence of environmental variables (11 variables chosen: oxygen OXI; temperature, T; coarse sand CS; salinity SALI; pH; coarse elements CE; organic matter OM; porosity PORO; fine sand FS; SILT and CLAY). Samples are also drown; the first and last digits refer to the sampling station (see Fig. 1), and the intermediate one to the sampling season (1=February samples; 2=March, 3=June, 4=September, 5=December).

be in relation to life histories of dominant species (Palacio et al., 1993; Carrasco & Carbajal, 1998), as in the sandy community from Plentzia where oscillations in density of suspension-feeders, mainly *Cerastoderma edule*, involve changes in feeding diversity. Seasonal variation in feeding structure is not always evident (Maurer & Leathem, 1981; Muniz & Pires, 1999), but a general pattern of variation of feeding diversity was observed in Gernika, basically due to an increase of the surface deposit-feeders density in September. Variation of this index in Plentzia and La Arena depended on the estuary and the different communities; nevertheless a decrease of feeding diversity in September was commonly observed. Other biological factors can also influence the structure of the benthos as bioturbation (Grehan et al., 1994).

We have compared our results to those obtained in other estuarine and marine areas with organic enriched environments, in order to find possible changes in feeding structure that could be assigned to a certain degree of disturbance. Figure 7 shows the feeding types observed in three of these coastal areas: sublittoral stations located in a fishery harbour (San Vicente Bay, Chile; Carrasco & Carbajal, 1998) and near a salmon farm (Washington, U.S.A.; Weston, 1990), and two intertidal communities affected by a paper mill (Pontevedra, Spain; Palacio et al., 1993). Deposit-feeding type prevails in those areas; in the more organic enriched sediments subsurface deposit-feeders are clearly dominant. An increase in the proportion of deposit-feeders is also observed in long-term studies, relating to periods of eutrophication and of oxygen deficiency (Beukema, 1991; Fallensen, 1992). Intertidal soft-bottoms in the Plentzia and Gernika estuaries, independently of their organic matter content, are systems widely dominated by deposit-feeders. However, subsurface deposit-feeders percentages are much lower than those observed in Figure 7, even in areas affected by sewage inputs, as the clayey community of Gernika. Although they were positively correlated with the organic matter content of the sediment, this feeding type was not significantly related with the granulometry, being the main feeding type only in the coarse sand community of Plentzia which had low val-

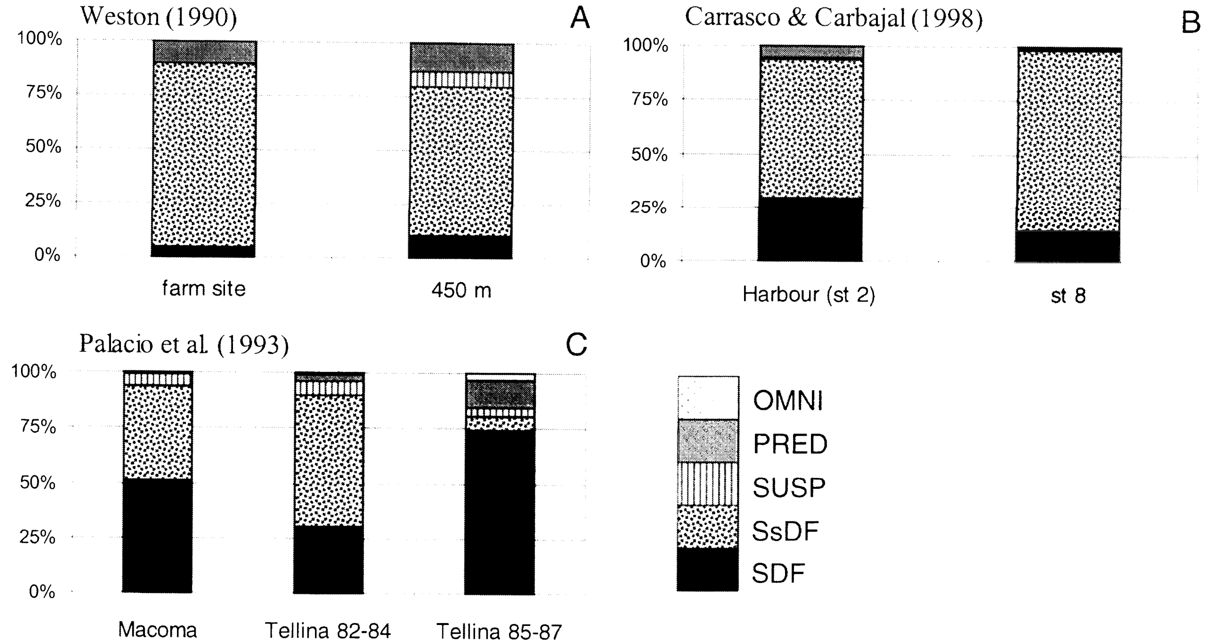

Figure 7. Relative abundance of feeding types observed in organic enriched environments (in the two first cases only polychaete fauna is considered). (A) Weston (1990). Feeding structure of the station closest to the farm site and of another one located far away. (B) Carrasco & Carbajal (1998). Sampling station 2 was located in the inner part of the bay, and usually was defauned; station 8 was located out of the harbour but presented a high organic matter content. (C) Palacio et al. (1993). The sampling station of the *Macoma* community was closest to the paper mill (average density from 1982 to 1987). *Tellina* community was less affected by the effluents (two periods are differentiated because a decrease of the harsh conditions of the sediment was observed).

ues of organic matter. This apparently contradictory relationship between subsurface deposit-feeders and sediment characteristics could be explained in part by the behaviour of some capitellid species, as *Capitella capitata* or *Heteromastus filiformis*. They are widely know as indicators of organic enrichment (Holte & Gullisek, 1987; Mendez et al., 1997), but we have found them in much lower densities than those of perturbed environments, and inhabiting different types of sediments, being considered as species of a wide ecological spectrum (García–Arberas, 1999). Other authors have also found apparently contradictory relationships between feeding types and granulometry. As with Cocito et al. (1990), we also expected to find more deposit feeders in sediments where mud predominated in La Arena estuary. The relatively low percentage of surface deposit-feeders and the absence of subsurface deposit species could be related to the quality of the sediments, as in this estuary the organic matter content of the sediment was much lower than expected because of their granulometry (García–Arberas, 1999). Further sediment analyses would be necessary in order to assess this possibility. However, subsurface deposit feeders were absent in areas with high levels of carbon available in the sediment (Bonsdorff & Pearson, 1999) and even a negative correlation between this feeding type and organic matter and silt and clay content is found (Muniz & Pires, 1999). The utility of feeding grouping approaches in detecting enrichment induced disturbances is limited and although organic enriched areas show a characteristic feeding structure, related to opportunistic species, a high dominance of subsurface deposit-feeders does not always mean an organic enrichment, as we have seen in the Plentzia estuary. Moreover, the separation of organic enrichment or pollution gradients from natural ones in these kinds of environments is not evident (Gray et al., 1988).

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'In vitro' and *'in situ'* decomposition of nuisance macroalgae *Cladophora glomerata* and *Pilayella littoralis*

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Key words: Pilayella, Cladophora, decomposition, invertebrate, nutrients

Abstract

The decomposition of two macroalgal species *Cladophora glomerata* (CHLOROPHYTA) and *Pilayella littoralis* (PHAEOPHYTA) was studied in the laboratory and field conditions. These species are known to cause the extensive 'macroalgal blooms' in the whole coastal range of the Baltic Sea. The objective of the experiments was to determine decomposition rates of the macroalgae, follow the changes in tissue nutrient content and validate the role of benthic invertebrates in this process. In the laboratory conditions, the differences in the decomposition rates of the algae were mainly due to the oxygen conditions. The weight loss of *C. glomerata* was slightly higher in anaerobic conditions than in aerobic conditions. If 99% of initial dry weight of *P. littoralis* was lost in aerobic conditions then only 20% was lost in anaerobic conditions. In general, the loss of phosphorus and nitrogen in algal tissues followed the weight loss. As an exception, the amount of nitrogen changed very little during the decomposition of *C. glomerata*. In field conditions, the photosynthetic activity exceeded the decomposition rate of *C. glomerata* at lower temperatures in spring. The decomposition of *P. littoralis* was estimated at 49% of its initial dry weight. The addition of benthic invertebrates had no effect on the decomposition process. In summer, the decomposition rates were estimated at 65% for *C. glomerata* and 68% for *P. littoralis* being in the same order of magnitude as observed in laboratory conditions. If the decomposition of *C. glomerata* was faster at the end of the experiment, the most significant losses of weight of *P. littoralis* took place during the first 2 weeks of deployment. *Idotea baltica* significantly contributed to the loss of *C. glomerata*. The decomposition rate of *P. littoralis* was reduced by the presence of *Mytilus edulis* and increased by *Gammarus oceanicus*.

Introduction

In the last three decades, the extensive supply of nutrients into coastal ecosystems has resulted in the excessive growth of filamentous macroalgae world-wide. As a consequence of large macroalgal 'blooms', the mass drift of algae is increasingly observed (Morand & Briand, 1996; Valiela et al., 1997). The development of algal mats pose both large-scale ecological problems (Norkko & Bonsdorff, 1996a,b; Valiela et al., 1997; Kotta et al., 2000) and economic damage (Ferreira et al., 1999). The algae, either attached or free floating, accumulate in very high biomasses (up to 400 kg wet weight m^{-2}) on the beaches, in lagoons, bays and estuaries (e.g. Briand, 1991). These algal mats can substantially modify nutrient dynamics both in a water column and sediment (Lavery & McComb, 1991; Peckol & Rivers, 1996). The decomposition of the algae often results in widespread anoxic conditions which, in turn, may seriously affect higher trophic levels and, hence, destabilise the whole shallow-water ecosystem (Norkko & Bonsdorff, 1996a,b; Hansen & Kristensen, 1997; Thiel et al., 1998).

A number of studies have documented increased production of filamentous algae in the Baltic Sea (Wallentinus, 1978; Mäkinen & Aulio, 1986; Vogt & Schramm, 1991; Bäck et al., 1991). These opportunistic species annually accumulate into unattached mats having maximum occurrence from July to September (Kautsky, 1995). The size of the algal mats reaches from a few square metres to a hectare; sometimes they exceed 0.5 m in thickness (Norkko & Bonsdorff, 1996a,b). Hence, the loose-lying algal mats constitute an important sink of nutrients in the coastal ecosystem of the northern Baltic Sea. The knowledge about the decomposition rate of the mat-forming macroalgae is essential to validate their role in the nutrient dynamics in the coastal ecosystems. Experimental studies quantifying the decomposition rate of benthic macrovegetation are scarce and usually they are carried out with vascular macrophytes (e.g. Pellikaan, 1984; Sfriso & Macromini, 1998).

The aim of this paper was to estimate the decomposition rate of the prevalent mat-forming filamentous macroalgae *Cladophora glomerata* (L.) Kütz and *Pilayella littoralis* (L.) Kjellm. in the coastal sea of the Gulf of Riga, northern Baltic Sea. Both *in situ* and laboratory experiments were conducted to investigate the effect of temperature, algal species and benthic invertebrates (herbivores, detrivores and filter-feeders) on the rate of algal decomposition and release of nitrogen and phosphorus. The field experiments enabled us to estimate the seasonal differences in the decomposition rate of macroalgae and to validate the role of benthic invertebrates in this process. The laboratory studies were performed in summer conditions only in which the algal decomposition rates and the release of nutrients were measured at aerobic and anaerobic conditions.

Materials and methods

Macroalgal decomposition experiments were performed with brown algae *Pilayella littoralis* and green algae *Cladophora glomerata* in laboratory and field conditions in spring and summer 2000. In the laboratory, the freshly collected algae (about 20 g wet weight, i.e. 3 g dry weight) with no macroepiphytes were placed into nylon mesh bags of 5.5 cm diameter, 20 cm height and 1 mm mesh size. These bags were incubated in 3 l flasks containing natural seawater at 18 °C. The water used in the experiment was screened through 55 μm mesh.

The incubation was run in aerobic and anaerobic conditions in the dark to keep the growth to a minimum. In aerobic conditions, the oxygen content was kept between 8.1 and 8.4 mg l^{-1} using aeration (1500 cm^3 min^{-1}). Twice a week the water in the incubation flasks was changed to avoid high nutrient concentrations in water. In the anaerobic conditions the flasks were closed with stopper and water was not changed.

The incubation lasted 35 days. Every 3–4 days, six bags of decomposing algal material (3 aerobic, 3 anaerobic) were analysed for dry weight and nutrients. Algal material was dried at 60 °C for 24 h. The content of total nitrogen and total phosphorus in remaining algal material was analysed using standard methods (Grasshoff, 1976; Solorzano & Sharp, 1980; Raimbault & Slawyk, 1991).

Field studies were carried out in Kõiguste Bay, northern Baltic Sea on 26.04.–25.05.2000 and 25.07.–24.08.2000. The water temperature ranged between 11 and 16 °C during spring and between 16 and 22 °C during summer. The algae were handled as described earlier. Nylon mesh bags with algal material were incubated at the depth of 2 m (anchored on a rope about 20 cm from the bottom). Besides algal material, some treatments included benthic invertebrates. We used the prevalent herbivore *Idotea baltica* (Pallas) (5 individuals per bag), the detrivore *Gammarus oceanicus* Segerstårle (5 individuals per bag) and the filter-feeder *Mytilus edulis* L. (10 individuals per bag) in our experiment. The incubation lasted 28 days. Once in a week, three replicates of each treatment were collected for determination of algal decomposition rate. For estimating the physiological state of algal material on different phases of decomposition, the net photosynthetic rates were measured using oxygen method. Comparative measurements with naturally growing algae in the study site were run in parallel. About 0.05 g (dry weight) of algal material were incubated in 600 ml glass bottles, filled with the sea water and incubated horizontally on special trays at 0.5 m depth. Bottles without algae served as the controls. The changes in dissolved oxygen concentration were measured by oxygen meter OXI 92 (Kotta et al., 2000).

Results

Laboratory experiments

The studied macroalgal species had different decomposition rates. However, these values varied highly depending on the conditions of incubation (Fig. 1). In aerobic conditions, *C. glomerata* lost about 46% of its initial dry weight after 5 weeks of deployment. The weight loss was linear at 1.4% per day (Fig. 1a). In anaerobic conditions the weight loss was 56% of initial dry weight with rapid weight loss of about 30% during the first week of decomposition (Fig. 1b).

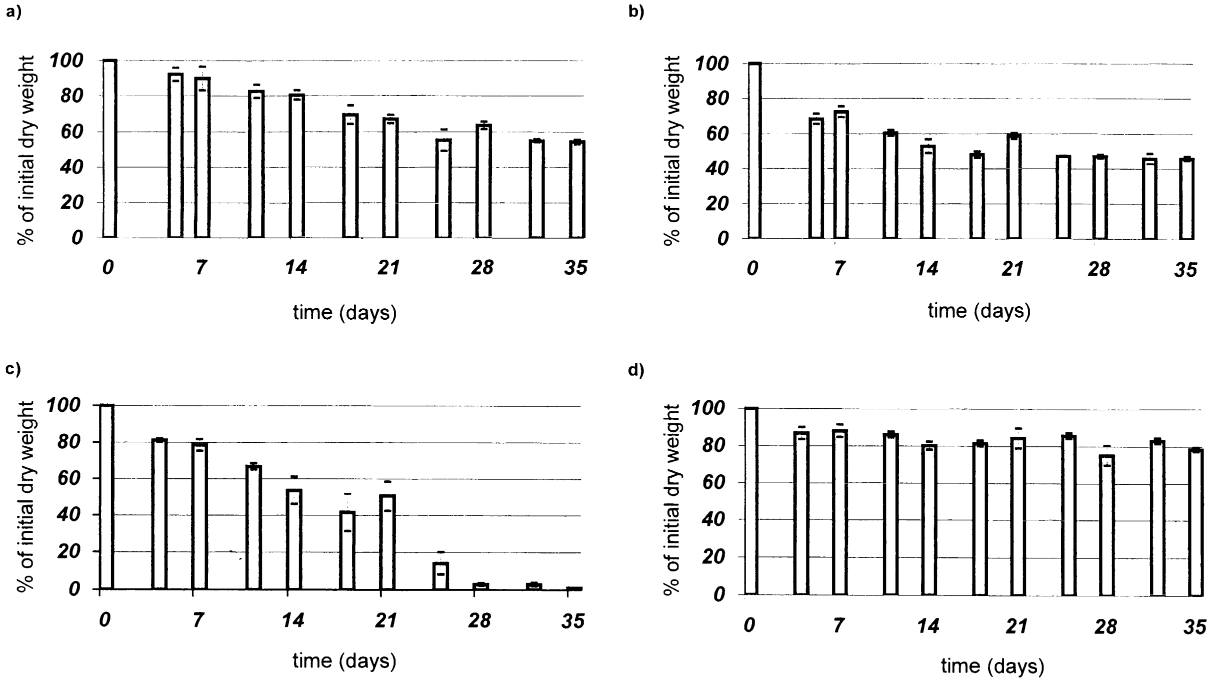

Figure 1. Decomposition of *C. glomerata* in (a) aerobic, (b) anaerobic conditions and *P. littoralis* in (c) aerobic, (d) anaerobic conditions (expressed in terms of per cent remained algal dry weight relative to initial dry weight±standard error, n=3).

Compared to *C. glomerata*, *P. littoralis* had an extremely rapid decomposition rate in aerobic conditions. During the experimental period, 99% of initial dry weight was lost at a linear rate of 2.8% per day (Fig. 1c). In anaerobic conditions, only 20% of the material was lost and similarly to *C. glomerata* the weight loss was most pronounced during the first week of decomposition (Fig. 1d).

As a rule, the losses of nitrogen and phosphorus in the decomposing material followed the trend similar to dry weight loss (Figs 2–5). *C. glomerata* lost only an insignificant proportion of total nitrogen pool in aerobic conditions (Fig. 2a). The nitrogen pool in the decomposing material decreased by 50% in anaerobic conditions (Fig. 2b). After 5 weeks of decomposition, only 8.3% of initial nitrogen pool had remained in *P. littoralis* in aerobic conditions (Fig. 3a), whereas no loss occurred in anaerobic conditions (Fig. 3b). Here the apparent increase in nitrogen pool may be attributed to the processes of nitrogen fixation.

In aerobic conditions nitrogen concentrations gradually increased both in *C. glomerata* and *P. littoralis* (Figs 2c and 3c). In anaerobic conditions, the concentrations remained almost unchanged in *C. glomerata* and increased in *P. littoralis* (Figs 2d and 3d). However, the later changes were considerably slower than in aerobic conditions.

In aerobic conditions, the phosphorus release of *C. glomerata* may be divided into two phases; phase I (days 0–14) in which there was a little decrease in phosphorus and phase II in which there was a relatively slow phosphorus loss. A total of about 40% of the tissue phosphorus was liberated during the decomposition process (Fig. 4a). In anaerobic conditions, a rapid loss of phosphorus occurred during the first 2 weeks of deployment in which phosphorus pool decreased by 50%. It was followed by a period of slower phosphorus liberation. At the end of incubation, about 25% of initial phosphorus had remained in the decomposing material (Fig. 4b). The phosphorus concentration of *C. glomerata* remained practically unchanged in aerobic conditions and decreased in anaerobic conditions. The most significant changes occurred during the first 7 days of incubation (Fig 4c and d).

In aerobic conditions *P. littoralis* released about 40% of phosphorus during the first week followed by a slower phosphorus liberation. Most tissue phosphorus was liberated at the end of incubation (Fig. 5a). In anaerobic conditions phosphorus was very rapidly released during the first 3 days in which the loss was about 50% of the initial value. Further on, no remarkable changes were observed (Fig. 5b). In aerobic conditions, the concentration of phosphorus showed a slow initial loss after 21 days followed by a rapid

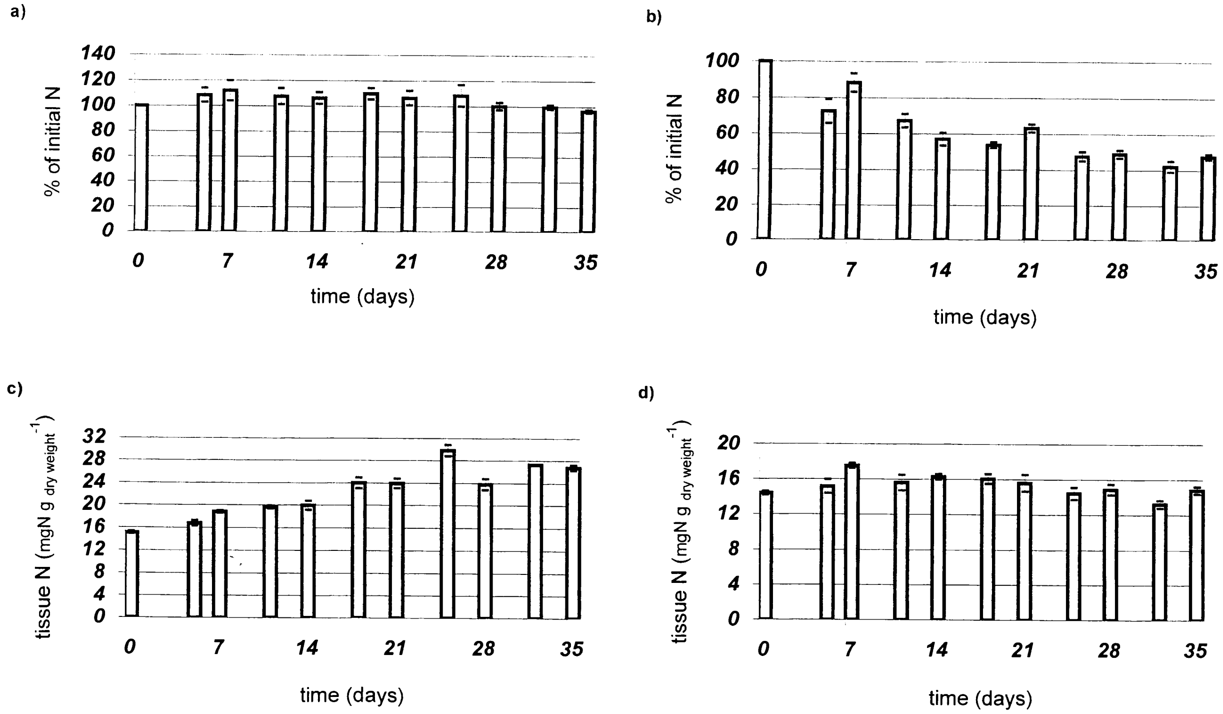

Figure 2. Total nitrogen in decomposing *C. glomerata*: changes in total nitrogen pool of algal material in (a) aerobic, (b) anaerobic conditions (expressed as per cent of initial nitrogen±standard error, *n*=3) and the changes in tissue nitrogen concentration in (c) aerobic, (d) anaerobic conditions (±standard error, *n*=3).

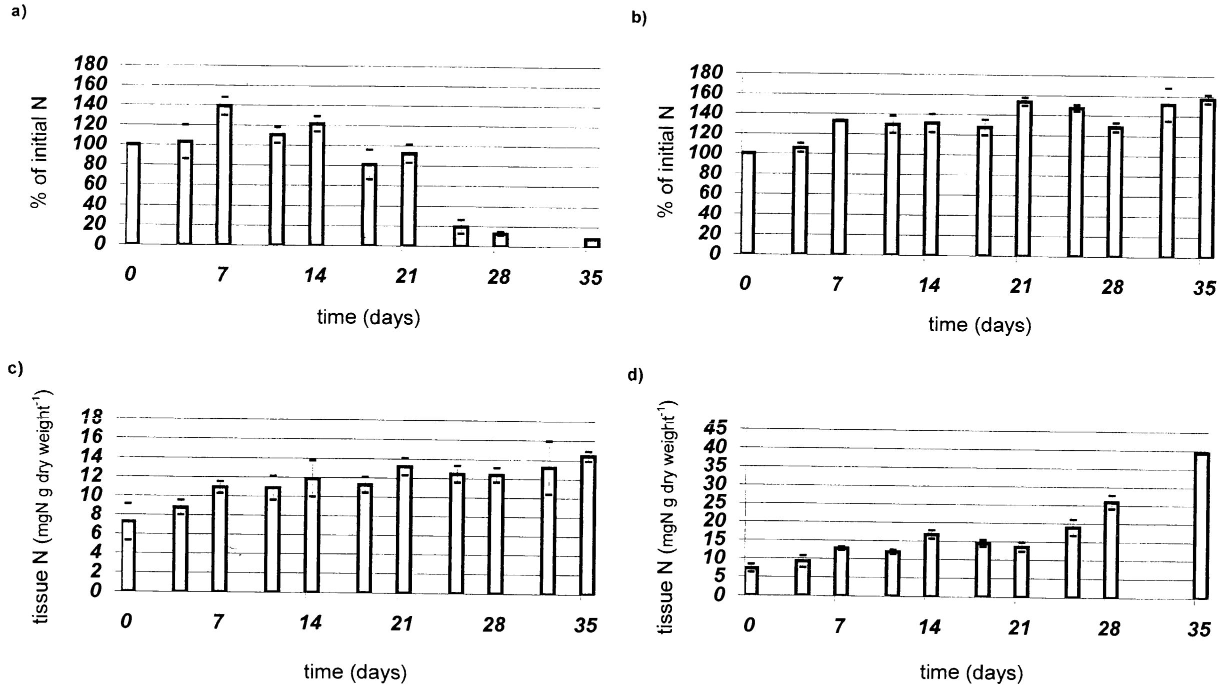

Figure 3. Total nitrogen in decomposing *P. littoralis*: changes in total nitrogen pool of algal material in (a) aerobic, (b) anaerobic conditions (expressed as per cent of initial nitrogen ±standard error, *n*=3) and the changes in tissue nitrogen concentration in (c) aerobic, (d) anaerobic conditions (±standard error, *n*=3).

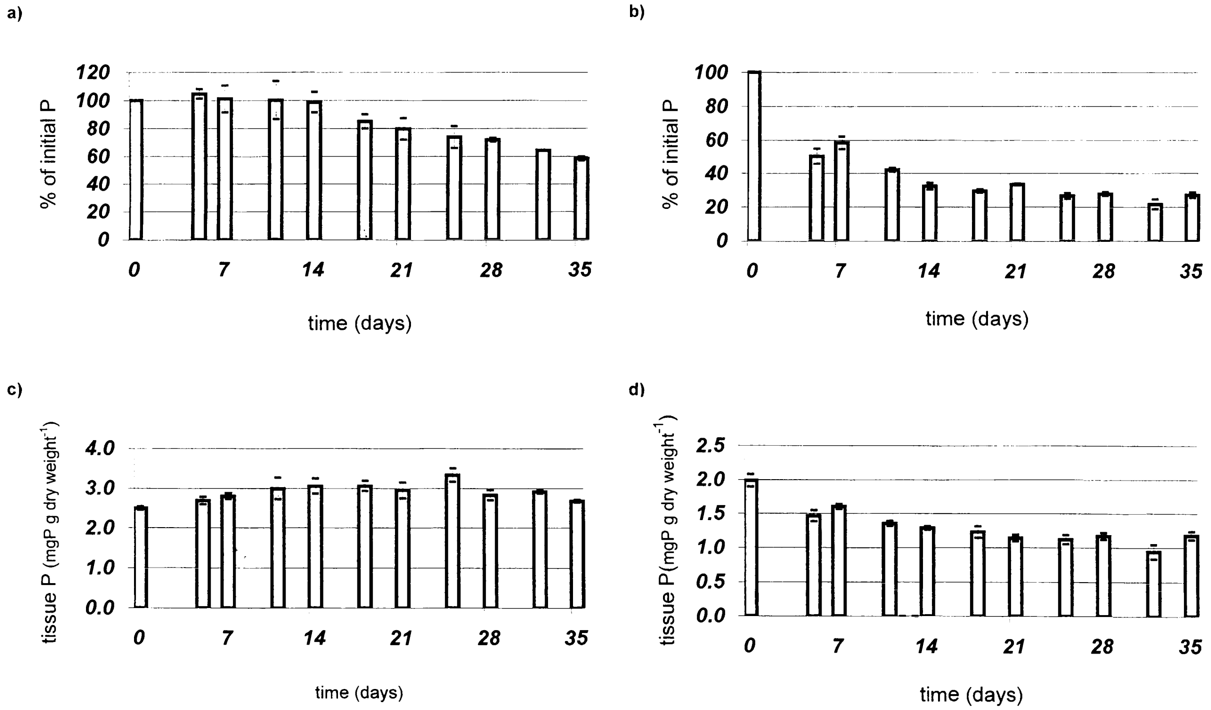

Figure 4. Total phosphorus in decomposing *C. glomerata*: changes in total phosphorus pool of algal material in (a) aerobic, (b) anaerobic conditions (expressed as per cent of initial phosphorus ±standard error, *n*=3) and the changes in tissue phosphorus concentration in (c) aerobic, (d) anaerobic conditions (±standard error, *n*=3).

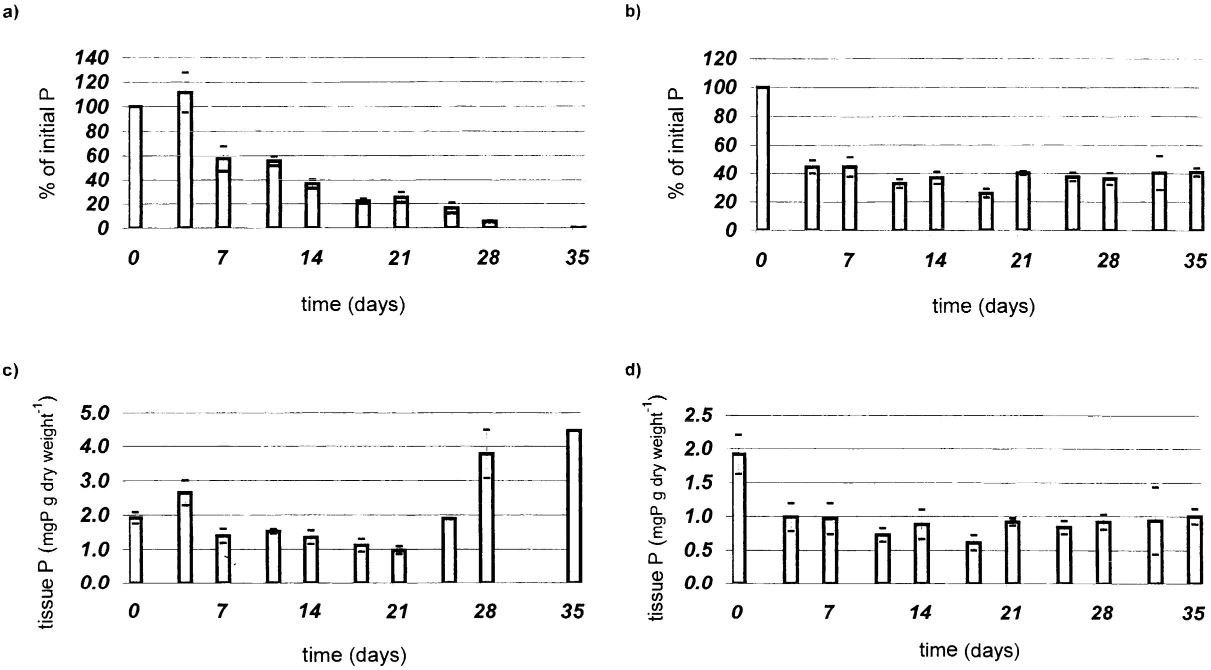

Figure 5. Total phosphorus in decomposing *P. littoralis*: changes in total phosphorus pool of algal material in (a) aerobic, (b) anaerobic conditions (expressed as per cent of initial phosphorus ±standard error, *n*=3) and the changes in tissue phosphorus concentration in (c) aerobic, (d) anaerobic conditions (±standard error, *n*=3).

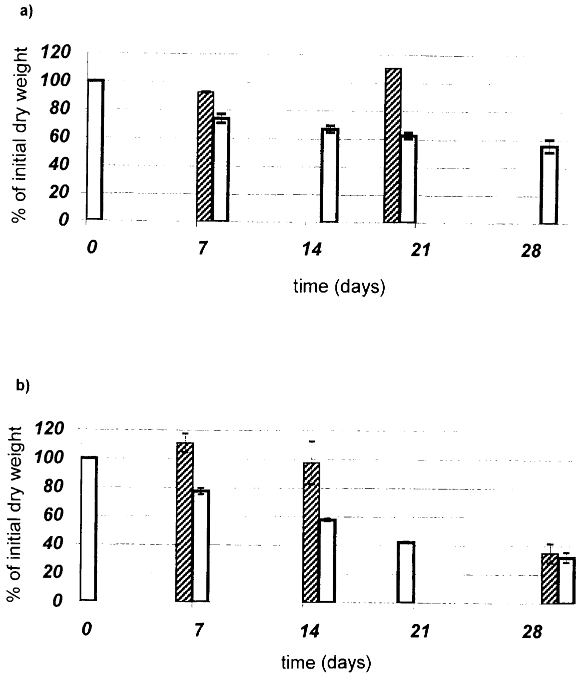

Figure 6. *In situ* changes in the dry weight of *C. glomerata* (hatched columns) and *P. littoralis* (white columns) in (a) spring and (b) summer (expressed in terms of per cent remained algal dry weight±standard error, n=3).

increase (Fig. 5c). In anaerobic conditions a rapid phosphorus decline during the first 3 days followed by stable values till the end of the experiment (Fig. 5d).

Field experiments

During spring, the dry weight of *C. glomerata* increased about 11% during 28 days of incubation. In the same period, *P. littoralis* lost about 49% of its initial dry weight (Fig. 6a). The net photosynthetic rates of *C. glomerata* were in average 25–30% of the values of naturally growing *C. glomerata*. *P. littoralis* showed no photosynthetic activity already after second week of incubation. The addition of invertebrates had no significant effect (one-way ANOVA, p>0.05) on the increment/decomposition rate of algae.

In summer, the decomposition rates were estimated at 65% for *C. glomerata* and 68% for *P. littoralis* (Fig. 6b). Net photosynthetic rates of *C. glomerata* were high during first weeks of incubation (up to 63% of the values of naturally growing *C. glomerata*). Low photosynthetic activity of *C. glomerata* was observed at the end of incubation. *P. littoralis* showed low photosynthetic activity only during the first week of the experiment. Herbivores had a significant effect (p<0.001) on the decomposition rate of *C. glomerata*. In these treatments, 89% of the initial weight were lost at the end of the incubation. Filter-feeders significantly (p<0.01) reduced the decomposition rate of *P. littoralis* during the first 7 days of incubation. If the algae alone lost about 22% of their initial dry weight then in the treatments with *M. edulis* only 14%, respectively. The effect of filter-feeders was insignificant at the end of incubation. This was probably due to hypoxic conditions, which were observed in the cages, inducing a significant mortality among filter-feeders. On the other hand, detrivores had a significant effect (p<0.001) on the decomposition rate of *P. littoralis* only at the end of the experiment inducing additional 18% dry weight loss of the algae (i.e. the total loss of 86%).

Discussion

The field experiments demonstrated that low temperature strongly inhibited both the decomposition process of studied algae and the activity of benthic invertebrates attached to these algae. Similarly to our findings, earlier studies (e.g. Carpenter & Adam, 1979; Birch et al., 1983) have demonstrated that temperature may be ranked among the most crucial factors regulating the decomposition process in field. However, it must be pointed out here, that our *in situ* experiments were performed with initially living algal material and at natural light conditions. Hence, at least at the early stages of the decomposition the photosynthetic processes are likely as important. The reason that *C. glomerata* did not loose its weight in spring incubation may, besides low water temperature, be attributed to remarkable photosynthetic activity of the 'decomposing' algae compensating at least partly the loss of algal degradation.

In summer, at higher temperatures, the decomposition of macroalgae was notably higher and the contribution of herbivores, detrivores and filter-feeders was significant. Living *Cladophora* is known to be very resistant to decay (Birch et al., 1983; Gabrielson et al., 1983). As compared to *Cladophora albida* in western Australia (Birch et al., 1983; Gabrielson et al., 1983), the decomposition of *C. glomerata*, however, is much faster in the study area. If *C. albida* lost only 20% of dry weight during 30 days, the value for *C. glomerata* was 65% (the further observations in our study area showed the loss of more than 99% of *C. glomerata* but also *P. littoralis* in 70 days of the incubation).

Nevertheless, a significant degradation of *C. glomerata* occurred only after 15 days of deployment. At the same time a remarkable decline of photosynthetic capacity of algal material was observed. As the fresh *C. glomerata* is not the preferred food for the adult *Idotea baltica* (Kotta, unpublished data) it may explain why the effect of *I. baltica* was significant only at the end of the experiment. By that time, the cell walls of *C. glomerata* were likely to be less resistant to herbivory (Birch et al., 1983) and the concentration of nitrogen and phosphorus was higher in the decomposing material making the algae more 'tasteful' for benthic invertebrates (Mann, 1988). The increases in tissue nitrogen and phosphorus concentrations as the decomposition progressed might most probably be attributed to microbial uptake or adsorption processes (Birch et al., 1983; Gabrielson et al., 1983; Buchsbaum et al., 1991; Pagioro et al., 1999).

Compared to *C. glomerata*, *P. littoralis* had a faster decomposition rate in aerobic conditions, whereas the species was found to be very resistant to decay in anaerobic condition. During the first 3 weeks of decomposition, the dry weight of *P. littoralis* decreased at the linear rate of about 3% per day both in laboratory (aerobic) and field conditions. Hence, the further decrease in the decomposition rate in field is likely to be caused by the onset of anaerobic condition. As the microbial activity in the decomposing material was not estimated, we can not qualify further whether the observed slow down of the decomposition was due to the biomass growth of micro-organisms.

During the first week of the experiment, the decomposition of *P. littoralis* was significantly inhibited by the presence of filter-feeding mussel *Mytilus edulis*. It is important to stress that we used initially alive algal thalli and due to photosynthetic activity growth took place together with decomposition process. *M. edulis*, owing to their large filtration activity and considerable biodeposition (Kotta, 2000), significantly increase the nutrient availability in the benthic system and hence, promote the growth of opportunistic *P. littoralis*. A similar advantageous effect of *M. edulis* has been previously documented also on the eelgrass *Zostera marina* (Reusch et al., 1994). *M. edulis* did not affect the decomposition rate of *P. littoralis* at the end of the experiment. The result may be attributed to the lower photosynthetic activity of the algae at the later stages of decomposition. Besides, followed by the development of hypoxic conditions in the decomposing material a significant mortality of *M. edulis* was observed in the second week of the experiment. The macroalgal mats have been acknowledged as a factor inducing widespread mortality of benthic invertebrates caused by temporal development of anoxic conditions during the decomposition process (Nicholls et al., 1981; Soulsby et al., 1982; Everett, 1994). As the photosynthetic activity of *C. glomerata* was low during the whole experimental period such a positive effect of mussels on the growth of algae was not observed.

Despite strong preference of *I. baltica* towards *P. littoralis* (Kotta et al., 2000), the effect of the herbivore on the algae was insignificant. This might be due to the toxicity of the algae (Lawrence et al., 1999) or hypoxic conditions inhibiting the activity of *I. baltica*. *Gammarus salinus*, which tolerates lower oxygen concentrations, significantly contributed to the degradation of *P. littoralis* at the end of the experiment.

A wide range of the rates of weight loss – from complete disappearance in a month to more than 2 years – has been reported for different species of macroalgae during decomposition. Because of differences in methods, experimental conditions and morphological nature, as well as the physiological state of algae, it is difficult to compare the values obtained by different researchers. Nevertheless, the decomposition rates measured in this study are in same order of magnitude as estimated for various macroalgae from different regions e.g. *Gracilaria tikvahiae*, *Delessaria sanguinea*, *Caulerpa cupressoides*, *Ulva lactuca* and *Laminaria sacharina* (Williams, 1984; Rieper-Kirchner, 1990; Buchsbaum et al., 1991).

The results of this study indicate that despite of differences in the decomposition of *P. littoralis* and *C. glomerata*, a remarkable amount of nitrogen and phosphorus are released from the decomposing algae already after 5 weeks of decomposition. Hence, the rapid decomposition provides an additional important supply of organic and inorganic compounds to be potentially available for recycling in the water. Released inorganic nutrients are likely to be assimilated by other macrophytes and phytoplankton, as the photosynthetic activity of detached mat-forming macroalgae is generally low. On the other hand, the increase in the nitrogen pool of *P. littoralis* (at anaerobic conditions) points to the possibility of immobilisation of nutrients at different stages of its decomposition either by microbial production or nitrogen adsorption processes (Buchsbaum et al., 1991). In such conditions, the decomposing algal mat could be regarded as an important sink of nutrients before the final degradation.

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Late Holocene pollution in the Gernika estuary (southern Bay of Biscay) evidenced by the study of Foraminifera and Ostracoda

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Key words: Foraminifera, Ostracoda, pollution, Gernika estuary, Holocene, palaeoenvironmental evolution

Abstract

The Arketas pier in the Gernika estuary (southern Bay of Biscay) has during summer periods waters with strong hypoxia (≤ 1 ml l^{-1}) but curiously bears the richest assemblages of foraminifers and ostracods in the whole estuary. The most abundant species in Arketas are: *Ammonia beccarii*, *Cribroephidium williamsoni*, *Haynesina germanica* and *Lobatula lobatula* (foraminifers) *Loxoconcha elliptica* and *Leptocythere castanea* (ostracods). The benthic foraminifer sub-species *A. beccarii tepida* exhibits in this estuary three different morphotypes, whose distributions are controlled by changes in the salinity, dissolved oxygen and nutrient content of the waters, and thus could be used as a good indicator of environmental alteration caused by those parameters. To determine whether this environmental alteration is due to natural causes or is anthropogenic, micropalaeontologic and sedimentologic contents of a sediment core 140 cm thick have been analysed. The study of Foraminifera and Ostracoda assemblages, species diversity, and *A. beccarii tepida* morphotypes allows us to reconstruct the evolution of this part of the estuary over the last 3800 years. To examine anthropogenic effects at this site, we compared the occurrence of heavy metals Pb, As, with the record of *A. beccarii tepida* morph C in Arketas. The maximum concentration of heavy metals coincides with the maximum occurrence of *A. b. tepida* morph C, in the 1970s (date calculated according to the estimated rate of sedimentation in this area). This indicates that distribution of *A. b. tepida* morph C is, at least partially, caused by heavy metal pollution. Nevertheless, the fact that we have found no deformed tests of this taxa in Arketas induces us to think that another factor, such as dysaerobia caused by eutrophization of nutrient inputs in several areas of this estuary, might have been responsible for at least of part of the anomalous shift of *A. b. tepida* morph C at that time. In recent times, an increase in this anthropogenic activity was evident, reaching its maximum in the 1970s and decreasing from the 1980s. During the late Holocene period (up to 3500 years BP), this site was a middle-outer estuarine settlement, euryhaline with low to moderate salinity. This episode ends with a marine transgressive pulsation. After depositional hiatus of unknown duration, recent sediments suggest a modern palaeogeographic position of Arketas located in a lower estuary environment. The study of *A. beccarii* through the Holocene sediments of Arketas indicates that dysaerobic/pollution conditions were not a singular event. At the same time, the increase of *A. b. tepida* morph C were particularly strong over several years of the 20th Century, at precisely the time that a noticeable decrease of richness and diversity of calcareous microfauna is detected in the estuary.

Introduction

Foraminifers are protists less than 1 mm in size, with pseudopodia and inner calcitic tests that can easily fossilize. They can thrive from the estuarine to the abyssal marine environments, on the bottom sediment (as benthics) and in the water column as planktonics. They have been known in geological records since the Cambrian (540 Ma, mega annum), and more than 40 000 species have been described (Loeblich & Tappan, 1988). Foraminifers are the most useful microfossils for dating and reconstructing palaeoenvironments of marine sedimentary rocks.

Ostracods are small (~1 mm) crustaceans with a bivalved, low-magnesium calcite carapace, which have been found in great numbers in marine and continental sedimentary rocks also since the Cambrian period. They are extremely useful in palaeoenvironmental analyses due to the rapid response of their assemblages to slight changes in the environment. Ostracods exhibit one of the most complete palaeontological records so far known, and more than 33 000 species have been described (Ostracon, 2000). Both foraminifers and ostracods are good indicators of salinity, dissolved oxygen, type of substrate and other environmental variables. We have today a reasonable knowledge of the distribution of many foraminifer and ostracod species as well as the main environmental factors controlling those distributions. These results have been used in the interpretation of past estuarine environments, on the assumption that the autoecology of these species has not substantially changed since late Tertiary times.

Early studies of foraminifers and ostracods from the southern Bay of Biscay were focussed to the knowledge of biocenosis composition, ecology and distribution of land/sea transitional species, pointing out the importance of main environmental factors responsible for microfaunal structure. Several estuaries have been described: Lekeitio (Pascual & Orue-Etxebarria, 1985), Gernika (Pascual, 1991, 1992), Bilbao (Rodríguez-Lázaro & Pascual, 1985; Pascual, 1991, 1992), Santoña (Cearreta, 1988) and San Vicente de la Barquera (Cearreta, 1989).

More recent works deal with Quaternary palaeoecology based on foraminifers, in the Bilbao estuary (Cearreta, 1998), the Bidasoa estuary (Cearreta, 1992, 1994) and the Santoña estuary (Cearreta & Murray, 1996); or based on foraminifers and ostracods, in the Gernika estuary (Pascual et al., 1998, 1999).

In particular, foraminifers and ostracods occurring today in the Arketas area of the Ria de Gernika estuary (Fig. 1) are characterised by assemblages where 2/3 species dominate. Despite the apparent monotony of the tests of dominant species, such as the foraminifer *Ammonia becarii* (Linnée), it is possible to discern interesting phenotypic variations on their morphotypes. The aim of this work is to show how these variations can be interpreted in terms of ecological and hydrological modifications and to describe these modifications in the Holocene geological record of the estuary. To do this, we first describe the microfaunal distribution and the phenotypes that currently live in this estuary; then we study their Holocene evolution along a sedimentary core in the Arketas area.

Description of sites studied

The Gernika estuary (Fig. 1) is located on the Bay of Biscay (43° 19′–43° 25′ N and 2° 39.5′–2° 41.8′ W) inside the Urdaibai basin, which extends for about 150 km^2; this basin was declared a protected area by the "Man and Biosphere Committee" of UNESCO in 1984. The estuary extends about 10.6 km in length and less than 1 km in width. The active channel has a depth averaging 2.6 m, though it can reach 4 m near the lower end of the estuary during the maximum tidal oscillation. The estuary runs through the town of Gernika, where it is possible to follow the tidal influence. It is oriented S–N and is geologically located on an anticline fold with faulted hinge, where plastic rocks of the Triassic Keuper outcrop. The Oca river is built on these materials and was invaded during the Holocene transgression.

The estuary has two types of substrate: (1) the upper estuary, which is formed by marsh of silty-clayey sediment colonised with *Spartina maritima* (Curtis) Fernald, *Phragmites australis* (Cav.) Trin. ex Stendel, *Salicornia ramosissima* J. Woods, *Puccinellia maritima* (Hudson) Parl., *Halimione portulacoides* (L.) Aellen and *Suaeda maritima* (L.) Dumort, (Benito & Onaindia, 1991), and (2) the lower estuary which is composed of sands deposited on the channel banks and beaches of the borders (Pascual, 1992). The boundary between the two zones is located along the Axpe-Kanala line (see Fig. 1). The tidal wave is the standard type as per Ketchum's (1983) terminology, but with specific characteristics in this area (Gobierno Vasco, 1986). The high tide seems to occur more or less at the same time in the whole estuary, but the low-tide point is displaced by the effect of lateral thinning at Axpe (Fig. 1). When the tide starts decreasing, this area is flooded and a great amount of water is retained; when it gets out, this water causes strong currents that mobilise the sediment. The water in Kanala is still flowing down two and half hours after the tide has started rising on the coast. Based upon foraminifer and ostracod assemblages, two main zones are distinguished in this estuary (Pascual, 1991, 1992; and Plate I). (1) The silty upper estuary has salinity ranging between 1 and 32‰; the foraminifer assemblage is dominated by *Ammonia beccarii* (Linné), *Haynesina germanica* (Ehrenberg) (as *H. depressula*), *Cribroelphidium wil-*

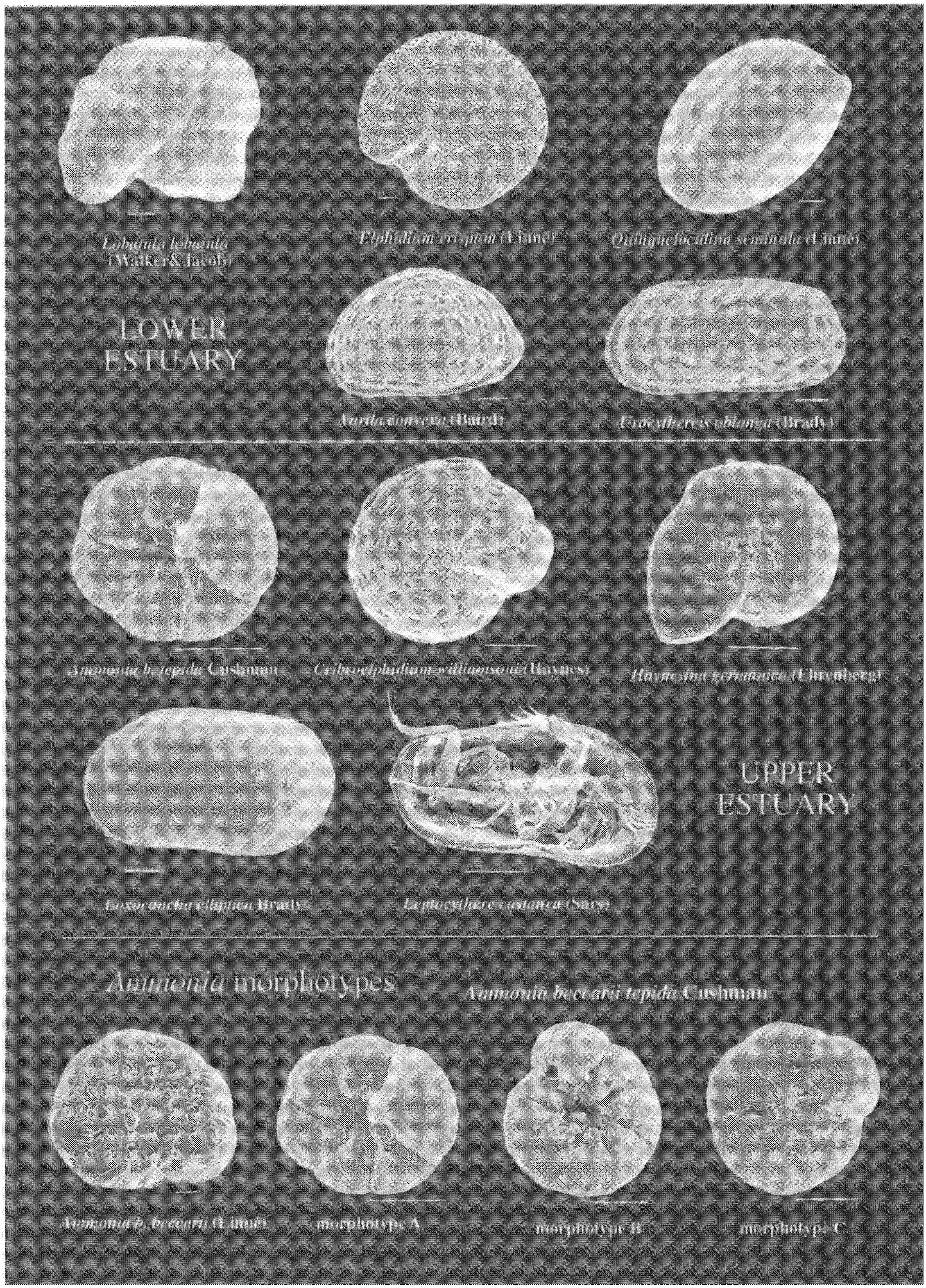
Lobatula lobatula (Walker&Jacob)
Elphidium crispum (Linné)
Quinqueloculina seminula (Linné)
LOWER ESTUARY
Aurila convexa (Baird)
Urocythereis oblonga (Brady)
Ammonia b. tepida Cushman
Cribroelphidium williamsoni (Haynes)
Haynesina germanica (Ehrenberg)
UPPER ESTUARY
Loxoconcha elliptica Brady
Leptocythere castanea (Sars)
Ammonia morphotypes
Ammonia beccarii tepida Cushman
Ammonia b. beccarii (Linné)
morphotype A
morphotype B
morphotype C

Plate I .

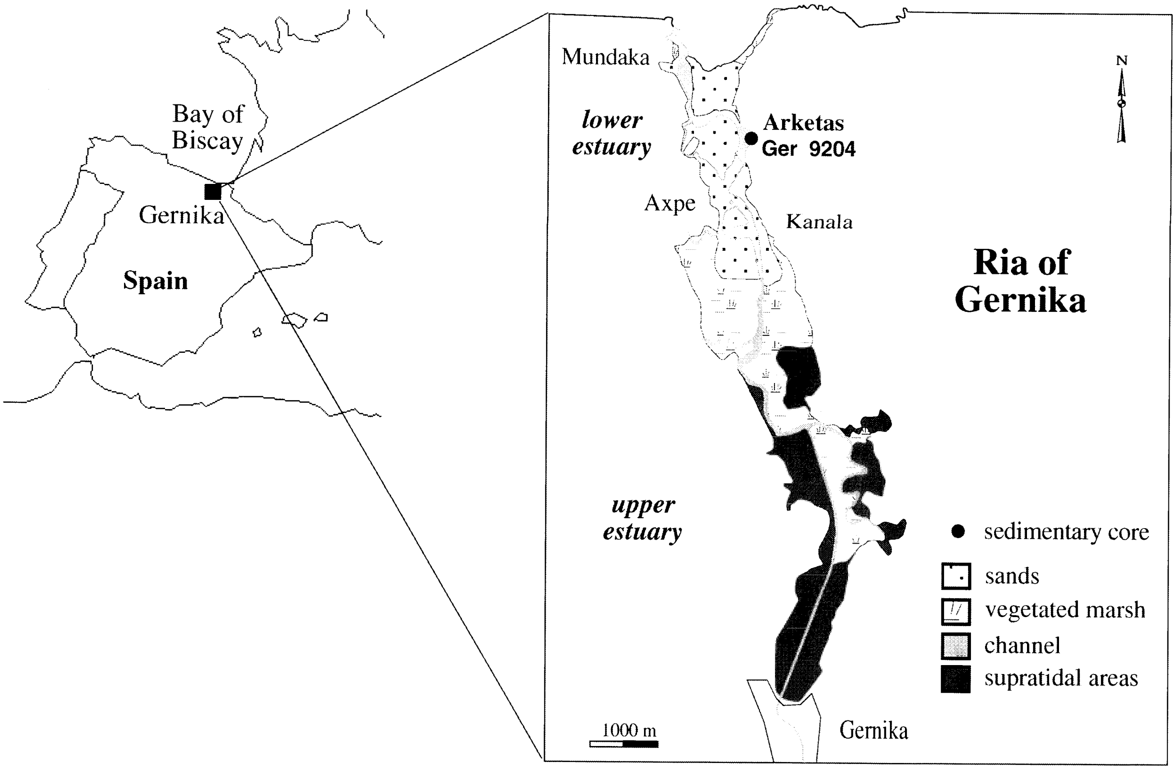

Figure 1. Location of site studied in the Gernika estuary.

liamsoni (Haynes) (as *Elphidium articulatum)* and the ostracods *Leptocythere castanea* (Sars) and *Loxoconcha elliptica* Brady. (2) Sandy lower estuary has marine character (S = 32–34‰) and the following assemblages: *Lobatula lobatula* (Walker & Jacob) (as *Cibicides lobatulus), Quinqueloculina seminula* (Linné), *Elphidium crispum* (Linné) (foraminifers) and *Urocythereis oblonga* (Brady), *Aurila convexa* (Baird) (ostracods).

Materials and methods

Recent foraminifers and ostracods from the Arketas pier were gathered from surface sediment with a Boltovskoy sampler. Samples were submerged in a methanol and Rose Bengal stain (Lutze, 1968) to differentiate living from dead specimens. A sedimentary core extracted by a Kullenberg sampler (140 cm of continuous record) was drilled in the Arketas area (location: 43° 23.8′ N–2° 4.8′ W) for palaeoecological analyses. Samples were taken every 10–20 cm, and completed in intervals with lithological and/or micropaleontological changes. Samples were washed, sieved (>63 μm) and around 300 individuals of benthic foraminifers were picked; counting was completed for planktonic foraminifers and ostracods. In several samples, a concentration of carapaces by flotation in heavy liquids (trichloroetylene) was necessary due to the scarcity of microfauna. Taxonomic analyses have been based on the Loeblich & Tappan (1988) (Foraminifera) and Athersuch et al. (1989), Guillaume et al. (1985) (Ostracoda) clasifications. Several microfaunal calculations were made: nF (No. of foraminifers per gram of dry sediment), nO (No. ostracods in 10 g of dry sediment), S (species richness), α index of Fisher et al. (1943), oceanity index (% planktonic/total foraminifers, Murray, 1976), and Shannon–Wiever diversity index for ostracods.

Sedimentologic study consisted in the description of lithology, texture, sedimentary structures, macrofauna, colour and cyclicity of the core material. A radiographic analysis (Baltograph) was performed in order to identify stratigraphic discontinuities, lithological changes and burrowing. Granulometrics (laser Malvern 2600 E difractometer) and carbonate content (Bernard calcimeter) were also obtained in the laboratory. A ^{14}C absolute dating from mollusc shells was performed by AMS (Accelerator Mass Spectrometry) at Beta Analytic In. in Florida (U.S.A.).

Ammonia beccarii *(benthic foraminifer) as environmental indicator*

A. beccarii (Linné) is one of the most widely studied species of benthic foraminifers, due to the morphological polymorphism of its tests, caused by changes in the environment. Sen Gupta et al. (1996) describe the occurrence of *Ammonia* in the continental shelf of the Northern Gulf of Mexico during summer periods with strong hypoxia. Karlsen et al. (2000) describe living specimens of *Ammonia* in the Chesapeake Bay (U.S.A.) as a facultative anaerobe tolerant of periodic anoxic conditions (DO = 0.47 and 1.72 ml l^{-1}). These authors relate deformed tests of specimens of this genus to strong modifications in benthic ecosystems.

A. beccarii is ubiquitous in the littoral marine to estuarine environment, and 22 different subspecies have been described (Ellis & Messina Catalogue, 1940–1974), though Schnitker (1974) considers many of those subspecies as ecophenotypic and juniors synonyms of *A. beccarii.*

A. beccarii has been used as a palaeoenvironmental indicator, taking into account the relative percentages of different morphotypes in the populations (Carbonel & Pujos, 1982). The particular points considered were: (1) shape and size of the umbilicus; (2) occurrence of umbilical plugs; (3) shape of umbilical lips and (4) the umbilical calcitic filling (see Plate I). The study of changes in dominance of different morphotypes of *A. beccarii* in Gernika allows us to follow environmental variations which are not detectable in classic micropalaeontological analyses.

Results

Foraminifers and ostracods currently living at the Arketas pier

A total of 22 species of foraminifers and 13 of ostracods have been found living in a silty-sandy sediment of the Arketas area during a year cycle (Table 1). During the summer, the lowest values of the dissolved oxygen were obtained (0.64 ml l^{-1}), with T = 22 °C and Salinity of 21‰. Under these conditions, only 11 species of foraminifers and 6 of ostracods live in Arketas during the summer time (see Table 1). The most important among the Foraminifera are *H. germanica* (29% of the sample), *L. lobatula* (18%), *C. williamsoni* (14%), *A. beccarii* (19%) and *E. crispum* (8%). The number of living specimens per gram of sediment is 88, and the Fisher's α diversity index is 2. Among the ostracods *L. elliptica* clearly dominates (67% of the sample), and *L. castanea* (12%), *Leptocythere pellucida* (Baird) (10%), *Semicytherura sella* (Sars) (8%), *Cytherois fischeri* (Sars) and *U. oblonga* as secondary components. The nO richness index is 70. This assemblage is comparable to the *Loxoconcha elliptica* assemblage described by Ruiz et al. (2000) as characteristic of euryhaline, highly polluted-muddy sediments from the southwestern Spanish estuaries.

Ammonia beccarii biocenosis in Arketas

At the Arketas pier, 2 subspecies of *A. beccarii* were detected: *Ammonia beccarii beccarii* (Linné) and *Ammonia beccarii tepida* Cushman. These subspecies are considered as species by different authors, and are named *A. beccarii* and *A. tepida*, respectively (Murray, 1991).

A. beccarii beccarii = Nautilus beccarii Linné. This is the bigger morph (length = 0.6 mm; width = 0.55 mm), with strong calcitic fillings of the umbilicus and sutures (see Plate I). It represents 10% of the living population of *Ammonia* in Arketas. Our specimens are comparable to those of *A. beccarii beccarii* on the Atlantic coast of France: Debenay et al. (1998: 238, Plate I, Fig. 3).

A. beccarii tepida = Rotalia becarii (Linné) var. *tepida* Cushman. Specimens are smaller in size (length = 0.35 mm; width = 0.30 mm) and weakly calcified. Three different morphotypes live today in the Gernika estuary (morphs A, B and C). Of those we only found in Arketas specimens belonging to the morphs B and C.

A. beccarii tepida form B. The umbilicus bears a plug of variable size (see Plate I) that in some cases completely closes the umbilicus. The average size is 0.35×0.30 mm. It accounts for up to 28% of the total living specimens of foraminifers in Arketas. This morph is similar to *Ammonia beccarii* from the north american eastern coast: Schnitker (1974: 216, Plate I, Figs. 5–6); *A.* gr. *beccarii* var. type *c* of Lac Tunis-Goulette, Tunis: Carbonel & Pujos (1982: 82, Plate I, Fig. 6); *A. tepida* morphotype 1 from Camarge, France: Pawlowski et al. (1995: 173, Fig. 3); *Ammonia parkinsoniana* (Orbigny) from the continental shelf of Louisiana: Sen Gupta et al. (1996: 228, Fig. 2B), and *A. parkinsoniana* from Chesapeake Bay: Karlsen et al. (2000: 501, Fig. 10, 2).

Table 1. Foraminifer and ostracod species living today in the Arketas pier. Number of individual tests (foraminifers) and valves (ostracods).

Summer	Spring	Winter	Autumn	Ostracods	Summer	Spring	Winter	Autumn	Benthic Forams
	2			*Aurila convexa* (Baird)	60	84	100	57	*Ammonia beccarii* (Linné)
		1		*Cushmanidea elongata* (Brady)		1	6	2	*Asterigerinata mamilla* (Williamson)
1	1	3	4	*Cytherois fischeri* (Sars)		1			*Bolivina pseudoplicata* Heron-Allen & Earland
	1			*Hemicytherura videns* (Mueller)				1	*Cassidulina crassa* d'Orbigny
		1		*Heterocythereis albomaculata* (Baird)	9	7	2	9	*Cribroelphidium lidoense* (Cushman)
6	2	1	8	*Leptocythere castanea* (Sars)	46	26	35	15	*Cribroelphidium williamsoni* (Haynes)
5	5		3	*Leptocythere pellucida* (Baird)		1	2	2	*Dorothia pseudotrochus* (Cushman)
	1			*Leptocythere tenera* (Brady)	26	12	41	6	*Elphidium crispum* (Linné)
35	5		10	*Loxoconcha elliptica* Brady				8	*Elphidium gerthi* Van Voorthuysen
	1			*Loxoconcha rhomboidea* (Fischer) Sars	1	2	4	3	*Elphidium macellum* (Fichtel & Moll)
		1		*Microcytherura fulva* (Brady & Robertson)	1				*Favulina hexagona* (Williamson)
4	4	1		*Semicytherura sella* Sars			2		*Gaudryina rudis* Wright
1	2	1		*Urocythereis oblonga* (Brady)	93	76	73	45	*Haynesina germanica* (Ehrenberg)
					59	74	111	34	*Lobatula lobatula* (Walker & Jacob)
								2	*Neoeponides mira* (Cushman)
						1			*Planorbulina acervalis* Brady
						1			*Quinqueloculina laevigata* d'Orbigny
								6	*Quinqueloculina lata* Terquem
						5	1	2	*Quinqueloculina seminula* (Linné)
					9	9	3	9	*Rosalina globularis* d'Orbigny
					2	1		2	*Textularia sagittula* Defrance
					13		29		*Triloculina oblonga* (Montagu)

A. beccarii tepida form C lacks umbilical plugs. The umbilicus is more or less filled by the extension of the umbilical lips, which may cover it completely. Size is 0.30 × 0.30 mm. In Arketas it accounts for 61% of living specimens of *Ammonia*. This morph is comparable to *Ammonia beccarii* variety *tepida* F3 generation from Delaware stock, from the north american eastern coast: Schnitker (1974: 216, Plate I, Fig. 1), and to *A. beccarii* var. *tepida* morph b of Lac Tunis-Goulette, Tunis: Carbonel & Pujos (1982: 82, Plate I, Fig. 5).

Geological evolution

In order to decipher the recent geological evolution of this area, the lithology and micropalaeontology of sedimentary core Ger 92-04 were studied.

Description of core (Fig. 2)

Over a length of 140 cm this basically sandy core shows two thinning-upward sequences overlaid with a sandy layer. From the base (140 cm, core depth) to 90 cm, the first thinning-upward sequence occurs, in which fine grey sands are overlaid by sandy clayey laminae ending with mud bearing many gastropod shells (*Turritella*). This sequence has a low carbonate content (6,7% average). Level 100 cm has been dated as 3740±60 years BP.

At 90 cm, a sharp contact marks the base of another thinning-upward sandy sequence up to 10 cm. This sequence starts with a shell lag level, then it progressively changes to a fine sandy layer with shell debris and plant fragments. Only the last 2 cm of this sequence are made of clay sediment. The carbonate content increases substantially in this interval, reaching its maximum for the series (48%) in its lower part. The remaining 10 cm up to the core top is composed of medium to fine sand with much shell debris. Carbonate in this level averages 18%.

Calcareous microfauna

A total of 41 species of benthic Foraminifera and 29 of Ostracoda have been identified (Tables 2–4). Based

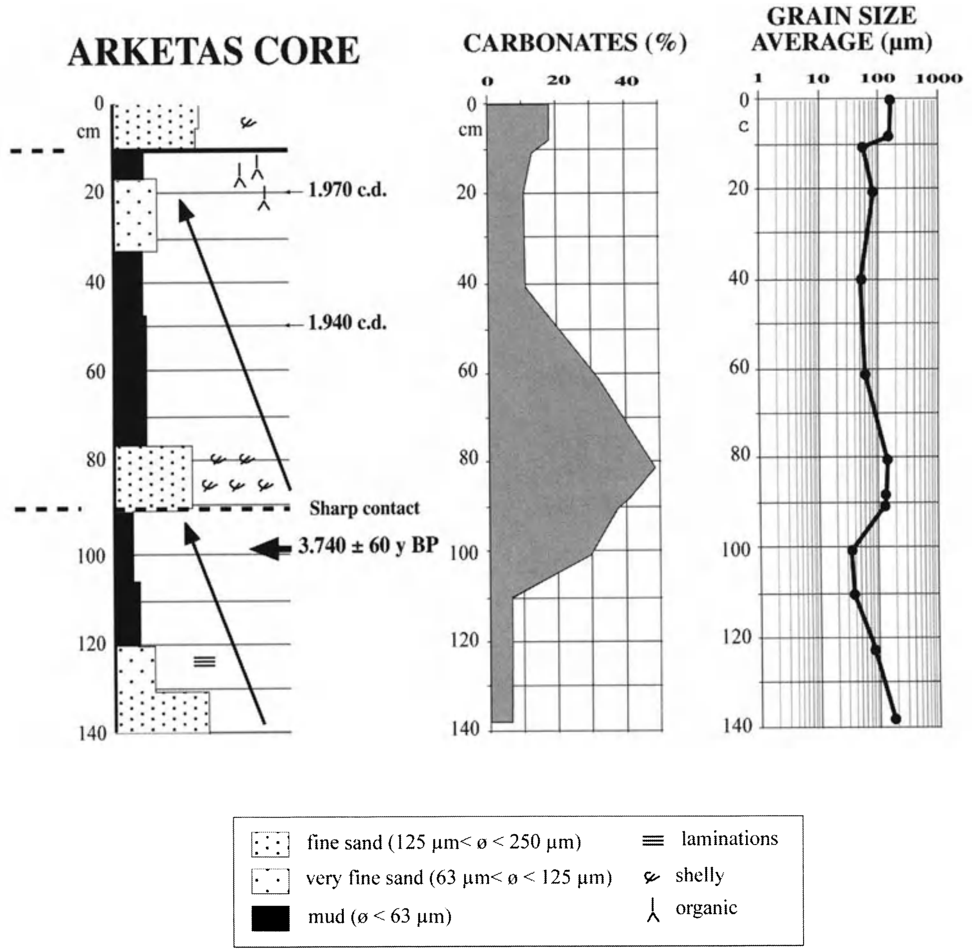

Figure 2. Lithology, carbonate content and granulometry of Arketas sedimentary core. Age estimations based on ^{14}C absolute dating (3740±60 years BP) and on the sedimentation rate obtained by decrease of isotope ^{210}Pb exc. (20th Century calendar dates). See text for details.

on microfaunal content, three different units can be differentiated (Fig. 3):

Unit 1 (140–90 cm). Foraminifer tests belong mostly to the hyaline type (>95%). The dominant assemblage comprises the euryhaline species *A. beccarii tepida* (>50%) and *H. germanica* (up to 14%). Species richness averages 12 species per sample and the α index oscillates between 1 to 4 (see Fig. 3 and Table 4). The nF is low in the sandy base (up to 122), rising towards the muddy modern terms (up to 1237). Ostracods are only present in the upper mid of this unit. Their richness nO is less than 259, and diversity index $H(S)$ increases up to 2.63 at the top of the unit. Assemblages are dominated by the estuarine species *L. elliptica, L. pellucida* and *Leptocythere psammophila* Guillaume. *A. beccarii tepida* form B dominates in the base of this sedimentary unit (79%), while morph C reaches 14% in the base and *A. beccarii beccarii* is a minor component of these assemblages (up to 1%).

A third morph of *A. beccarii tepida* (form A) also occurs in these levels of the Arketas core. This morph is characterised by an open umbilicus and reduced umbilical lips with no plugs. Size is small (length = 0.25 mm; width = 0.20 mm; Plate I). *A. beccarii tepida* form A is present in this unit with percentages less than 7%. Individuals of this morph are comparable to *A. beccarii* forma *tepida* from Los Pensiquitos Lagoon, California: Walton & Sloan (1990: 133, Plate I, fig. 1b) and also to living specimens of *A. beccarii tepida* from Long Island, U.S.A.: Pawlowski et al. (1995: 173, Fig. 3, 2).

In modern levels of this unit, *A. tepida* form C increases its percentages (51%).

Table 2. Benthic foraminifer species studied in the Arketas core. Number of individuals

GER 9204 BENTHIC FORAMS		UNIT 1			UNIT 2				UNIT 3					
sample (cm, core depth)	total	140	120	110	100	90	87	80	60	40	20	10	8	2
Ammonia beccarii (Linné)	1264	29	98	277	226	81	118	117	109	47	48	19	17	9
Asterigerinata mamilla (Williamson)	76	1		2	1	22	12	12	19	2	3	1		1
Brizalina variabilis (Williamson)	19	1				9	3	1	5					
Lobatula lobatula (Walker & Jacob)	403	5	4	4	15	88	79	66	62	15	15	4	7	4
Cribroelphidium williamsoni (Haynes)	79	6	4	9	3	2	2		2	3		2		
Elphidium crispum (Linné)	124	2	6	14	21	8	11	9	10	2	1		1	7
Elphidium macellum (Fichtel & Moll)	4	1										1		
Globulina gibba (d'Orbigny)	2	1						1						
Haynesina germanica (Ehrenberg)	335	8	16	21	25	42	31	20	32	12	16	5		2
Massilina secans (d'Orbigny)	6	1					4							1
Quinqueloculina lamarckiana d'Orbigny	1	1									1	1		8
Quinqueloculina seminula (Linné)	37	1					3	2			1	1		8
Jadammina macrescens (Brady)	3			2						1				
Brizalina spathulata (Williamson)	7				1	1	2	1	2					
Rosalina globularis d'Orbigny	52				3	15	15	12	5	1	1			
Bolivina punctata d'Orbigny	2					2								
Bolivina pseudoplicata Heron-Allen & Earland	13					7	1	4	1					
Brizalina alata (Seguenza)	10					3	1		6					
Cassidulina obtusa Williamson	8					3	1	2	2					
Favulina hexagona (Williamson)	9					3	1	3		1				
Patellina corrugata Williamson	10					1		7	1	1				
Planorbulina mediterranensis d'Orbigny	2					1	1							
Planorbulina variabilis (d'Orbigny)	1					1								
Pyrgo williamsoni (Silvestri)	2					1	1							
Rosalina irregularis (Rhumbler)	12					2								
Triloculina oblonga (Montagu)	20					2								2
Uvigerina peregrina Cushman	2					1			1					
Brizalina subaenariensis (Cushman)	4						1	1	2					
Cyclogyra carinata (Costa)	1						1							
Elphidium gerthi Van Voorthuysen	7						1	5	1					
Lenticulina calcar (Linné)	1						1							
Quinqueloculina lata Terquem	3							1						2
Angulogerina angulosa (Williamson)	2							2						
Cribroelphidium lidoense Cushman	11						1		1					
Fissurina orbignyana Seguenza	1						1							
Lagena striata (d'Orbigny)	1						1							
Eponides repandus (Fichtel & Moll)	2							1						
Fissurina sp.	2							2						
Lagena sulcata (Walker & Jacob)	1							1						
Lepidodeuterammina ochracea (Williamson)	2								1					
Quinqueloculina laevigata d'Orbigny	1													1
no indv. per sample	2541	57	128	329	295	293	266	264	87	85	33	25	37	

Table 3. Ostracod species studied in the Arketas core. Number of valves.

GER 9204 OSTRACODS			UNIT 3				UNIT 2				UNIT 1			
sample (cm, core depth)	2	8	10	20	40	60	80	87	90	100	110	120	140	no. valves
Palmoconcha laevata (Norman)									4	1				6
Loxoconcha elliptica Brady							5	1	12	4				22
Basslerites berchoni (Brady)							2	2		1				5
Caudites calceolatus (Costa)				1		1	1	2		1				6
Loxoconcha rhomboidea(Fischer) Sars						3	2	5	11	1				22
Leptocythere pellucida (Baird)							3		7	2				12
Leptocythere psammophila Guillaume		2				2	1	4	4	2				15
Aurila convexa (Baird)	1					1		1	7					10
Neocytherideis subulata (Brady)							1		1					2
Cytherois fischeri (Sars)						1	2		1					4
Hemicytyherura videns (Mueller)				1		1	4		7					13
Semicytherura sella Sars						2	1	2	2					7
Xestoleberis aurantia (Baird)									2					2
Cytherura gibba (O.F. Muller)									3					3
Leptocythere castanea (Sars)							2	3	3					8
Leptocythere sp.									2					2
Paradoxostoma normani Brady									2					2
Aurila woutersi Horne				1			4	2	1					8
Callistocythere pallida (Mueller)					1	1	4	2	2					10
Hemicytherura sp.						1	1	1	1					4
Semicytherura sp.						1	1		3					5
Heterocythereis albomaculata (Baird)						2			1					4
Urocythereis oblonga (Brady)						2		1						3
Microcytherura fulva (Brady & Robertson)						2	1	1						3
Leptocythere lacertosa (Hirschmann)				1			2	1						2
Neocytherideis faveolata (G.W. Muller)						1								2
Semicytherura robusta Bonaduce et al.						1								1
Sarsicytheridea bradii (Norman)						2								2
Eucytherura sp.				4	1	1								1
no. indv. per sample	1	2	0			24	37	28	76	12	0	0	1	186

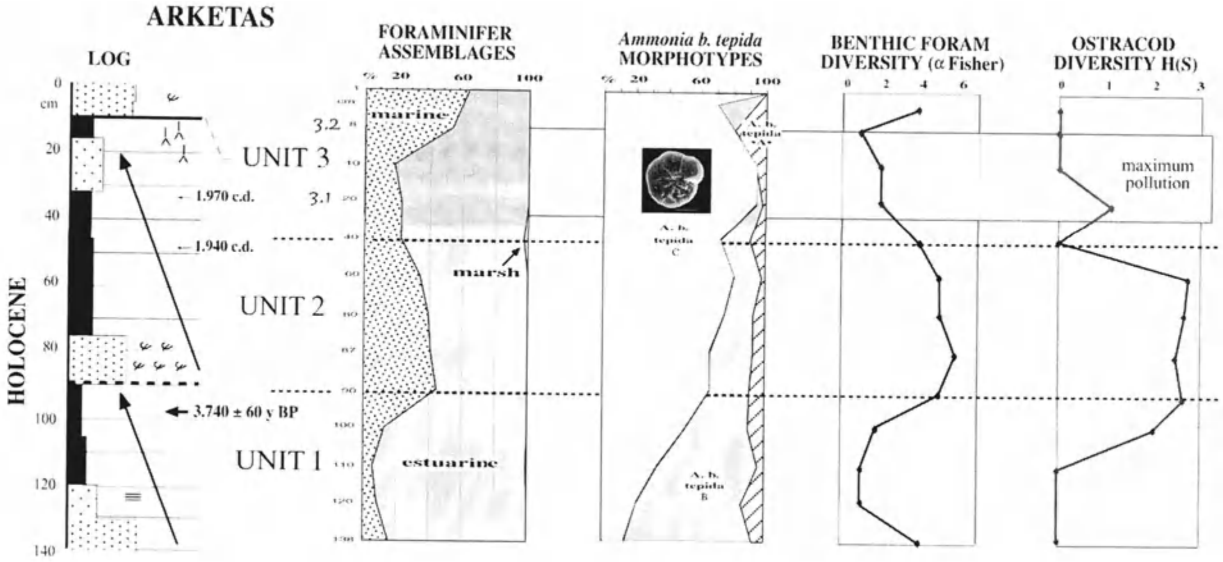

Figure 3. Distribution of benthic foraminifer assemblages, *Ammonia beccarii* morphotypes, and diversity indices of benthic foraminifers and ostracods in Arketas sedimentary core.

Unit 2 (90–40 cm). Diversity of benthic foraminifers increases (S = 19–23; α = 5–6; nF up to 1241, Table 4), and the foraminifer species found are, besides those described in the former unit, other characteristic of coastal areas: *L. lobatula* (25% of the sample), *Rosalina globularis* d'Orbigny and *E. crispum.* Other minor assemblage composed of marine shelf species *Bolivina pseudoplicata* Heron-Allen & Earland and *Cassidulina obtusa* Williamson, also occur. Ostracod assemblages are more diverse and include marine shelf species too: *L. rhomboidea*, *L. psammophila, Hemicytherura videns* (G.W. Müller), *Xestoleberis aurantia* Baird, *Sarsicytheridea bradii* (Norman) and *Eucytherura* sp. A noticeable number of specimens of planktonic foraminifers also occur in these levels (oceanity index <7). In this unit, form C of *A. b. tepida* dominates over morph B, increasing its dominance from bottom to top of the interval (66–81%). Correlatively, morphs B and A decrease along the same interval (25–17% and 9–2%, respectively). *A. b. beccarii* never reaches 3% of the total *Ammonia.*

Unit 3 (40 cm to core top). Assemblages with euryhaline species dominate this interval (*A. beccarii, H. germanica*), and some specimens of other species typical of the coastal shelf *(L. lobatula, Q. seminula*) also occur. Microfaunal richness and diversity are lower in this unit (nF up to 161; S = 4–12; α = 1–4; Table 4); ostracods are practically absent from these levels, with only scarce specimens belonging to species of coastal affinities. Two different sub-units can be distinguished. Sub-unit 3.1) (40–10 cm): assemblages are quite rich (nF: 132–161), *A. b. tepida* is well represented (72–94%), with its morph C being dominant (up to 94% of the sample). Sub-unit 3.2) (10 cm to top) richness decreases abruptly (nF < 44); *Ammonia* is scarce (<16 specimens/sample) and morph C is still dominant. Morph B, in turn, is very secondary and may be absent in some levels (see Table 4).

Discussion

Palaeoenvironment interpretation with microfaunal assemblages

Benthic foraminifers and ostracods have been used as indicators of environmental changes produced by human activity, particularly in transitional and coastal environments, where ecosystem alterations are provoked by metal and nutrient pollution (Alve, 1991; Pascual, 1991; Sharifi et al., 1991; Yanko et al., 1994; Pascual & Pujos, 1994; Geslin et al., 1998; Karlsen et al., 2000; Ruiz et al., 2000). *Ammonia* (Brunnich) is one of the most widely used genera in these environmental studies, due to the fact that some of its species are able to live in dysaerobic waters (Debenay et al., 1998).

The Arketas pier is, during summer time one of the least oxygenated areas of the estuary ($\leq$1 ml l^{-1}),

Table 4. Calcareous microfaunal (foraminifer, ostracod) diversity indices database in the Arketas sedimentary core.

GER 9204 MICROFAUNAL INDICES	BENTHIC FORAMINIFERS									OSTRACODS		
	sample (cm, core depth)	No individuals	nF (no individuals/g sample)	Number of species	Fisher's alpha index	Planktonics/total forams	*Ammonia b. tepida* morph A (%)	*Ammonia b. tepida* B (%)	*Ammonia b. tepida* C (%)	No individuals	Shannon-Wiever index H(S)	nO (no /10 g sample)
UNIT 3	2	37	36	10	4	2			100	1	0	9
	8	25	17	3	1	0	19	6	75	2	0	10
	10	33	44	7	2	0	6		94	0	0	0
	20	85	132	7	2	2	2	4	94	4	1.10	76
	40	87	161	12	4	0	9	19	72	1	0	18
UNIT 2	60	264	611	19	5	7	2	17	81	24	2.75	733
	80	266	414	19	5	4	7	18	75	37	2.67	578
	87	293	426	23	6	4	7	27	66	28	2.48	389
	90	295	545	21	5	7	9	25	66	76	2.63	1241
UNIT 1	100	295	567	8	2	1	10	39	51	12	1.79	259
	110	329	1237	7	1	0	4	62	34	0	0	37
	120	128	122	5	1	0	14	65	21	0	0	0
	140	57	63	12	47	0	7	79	14	1	0	11

but has the richest benthic foraminifer assemblages. This pier is located in a sheltered area inside the region of strong marine influence. The sedimentation rate is also high due to circular currents, to which is added the income from a nearby tributary; all of these factors produce a strong sedimentation of fine materials with high organic content and low redox potential (Gobierno Vasco, 1986). This is the cause of a strongly reduced environment (<−200 mV), with more than 5% of organic matter in sediments with clay granulometric content ranging between 40% and 90%. Organic matter is added to the pier from several sewage pipes.

Several authors (Boltovskoy, 1965; Carbonel & Pujos, 1982) have pointed out the low occurrence of benthic foraminifers with low oxygen conditions. Where anoxia occurs the richness, diversity and morphology of foraminifer tests undergoes strong modifications (Pujos, 1976; Sen Gupta et al., 1996; Karlsen et al., 2000). In turn, where the low oxygen and abundant organic matter co-occur, opportunistic species of Foraminifera and Ostracoda dominate the assemblages (Sen Gupta & Machain-Castillo, 1993; Ruiz et al., 2000). This happens in Arketas today, where the calcareous microfauna assemblages are dominated by opportunistic species *A. b. tepida* and *L. elliptica*.

A study of relative abundance of microfaunal species from a sedimentary core in Arketas shows palaeoenvironmental changes. We have analysed the relative percentages of foraminifer species characteristic of marine, estuarine and marsh environments, the relative percentages of *A. b. tepida* morphotypes, Fisher's benthic foraminifer α index , and the Shannon–Wiever $H(S)$ ostracod diversity index in the Arketas core (Fig. 3).

The occurrence of *H. germanica* and *A. b. tepida* in unit 1 indicates an estuarine, tidal, shallow milieu with vegetation and low salinity (Murray, 1991). Ostracods only appear in the upper part of this unit, with typical estuarine species of the genera *Leptocythere* and *Loxoconcha* (Guillaume et al., 1985; Athersuch et al., 1989; Pascual & Rodriguez-Lazaro, 1996; Ruiz et al., 2000). The ostracod diversity index $H(S)$ and the α foraminifer index increase towards the top of this unit, showing some stabilisation of the environment, very likely due to the progressive incoming of marine conditions in this area. Dominance of morph B in this interval (up to 79%, at the base of unit) points to the presence of euryhaline, oxygenated waters in this estuarine area (Carbonel & Pujos, 1982).

The sharp contact between units 1 and 2 (at 90 cm, core depth) indicates that sediments of unit 2 were deposited with no temporal continuity with those of unit 1. This erosive contact leads us to think that an unknown thickness of sediment is missing in this series, because it has been eroded by energetic currents. Dating of level 100 cm core depth as 3740±60 years BP suggests that the erosive level (90 cm) could be the consequence of a marine transgressive pulsation 3500 years ago in the Bay of Biscay (Pirazzoli, 1991; Pascual et al., 1998). In the deposition of Unit 2, diversity indices of foraminifers and ostracods (α, S(H), see Fig. 3) are the highest of the series, and decrease to the top of this unit, indicating some environmental alteration. Foraminifer assemblages, bearing species typical of marine shelves (*B. pseudoplicata*, *C. obtusa*, *L. lobatula*, *R. globularis*, *E. crispum*), marine shelf ostracods species like *S. bradii*, *Eucytherura* sp., and the occurrence of a high number of planktonic foraminifers (oceanity index = 7), clearly show the entry of open marine waters in several levels of this unit. Thus the palaeogeographic emplacement of Arketas during that time was that of an outer estuary with strong open marine influences. Such marine influx has previously been described in other areas of the Gernika estuary (Pascual et al., 1998). As a consequence of this, the Arketas pier was a less confined and more open milieu than in the former unit. This is also supported by the morphology of *A.* b. *tepida* that shows the dominance of morph C over B (see Fig. 3). Morph C is a good indicator of degraded environments (Seiglie, 1975; Pascual, 1990). In the Bilbao estuary, described as one of most polluted estuaries in Europe, this morph dominates the foraminiferal assemblages (Pascual, 1990). Morph C is also adapted to strong salinity variations, included those of inner shelf (Carbonel & Pujos, 1982), and is the morph that dominates in Arketas (51–81%) when salinity increases with the income of marine waters.

Two different phases of environmental evolution can be differentiated in Unit 3 (see Fig. 3). During the first one (3.1), the strong decrease of microfaunal diversity indices clearly points to an environmental change in Arketas. The maximum occurrence of *A. b. tepida* C in this sub-unit, coincident with a predominantly estuarine foraminifer assemblage and the scarcity of ostracod fauna, support the fact that the environmental alteration could be related to a factor other than salinity (e.g., strong decrease of dissolved oxygen in the water, pollution, etc.). A relative return to marine conditions is shown in sub-unit 3.2, suggested by very poor foraminifer assemblages with littoral species, and by the dominance of *A. b. tepida* C in modern levels of this core, evidencing the settlement of an outer estuarine environment in Arketas today.

Ammonia beccarii *and the environment*

Because the genus *Ammonia* is an euryhaline opportunistic taxa that may survive in dyxoxic environments, it is found as dominant in transitional euryhaline estuarine and coastal milieux. In some cases, specimens can show deformities in shell morphology due to metal contamination, shortage of nutrients and/or persistence of hypoxic conditions (Sen Gupta et al., 1996; Karlsen et al., 2000). The low concentration of oxygen is also evidenced by the dominance of *A.b. tepida* morph C, which is adapted to dysaerobic environments (Carbonel & Pujos, 1982; Tourte, 1987; Pascual et al., 1991).

A. b. tepida morph C occurs today in waters with strong salinity variations inside some estuaries (Carbonel & Pujos, 1982; Pascual et al., 1991). In the geological record of Arketas, we found that morph C dominates from the top of unit 1, coincident with the entrance of marine waters, that is, to an increase in salinity. These conditions are maintained throughout unit 2, but where an estuarine (euryhaline) regimen returns to this area during the deposition of sub-unit 3.1, the maximum of morph C occurs, which indicates that another parameter besides salinity is controlling the distribution of this morphotype.

A similar result was described in lake Tunis (Carbonel & Pujos, 1982), where the pH decreases and the oxygen is very scarce during the summer. In that lake, the *Ammonia* populations modify their morphology to produce the C morph, the best adapted to low-oxygen conditions. The persistence of hypoxia allows for the expansion of C morphotypes of *Ammonia*, which seem to prefer a milieu of sea-lagoon transition.

The same has been observed in Bay of Berre, west of Marseille, France (Tourte, 1987). There, a large number of specimens of *A. b. tepida* morph C were found in euryhaline areas with clay substrate. During summer and fall times, a regime of hypoxia (1.2 ml l^{-1}) settles in the bottom waters, due to the eutrophization of surface waters. The decreasing of pH acidifies bottom waters, which prevents the normal calcification of morphs B of *A. b. tepida*, as was confirmed in the laboratory by Tourte (1987). In Chesapeake Bay (U.S.A.), several authors have established that oxygen depletion occurs when the rate of benthic respiration exceeds the re-oxygenation rate during the spring and the strong dysoxia remains during the summer (Officer et al., 1984; Boynton & Kemp, 1985; Karlsen et al., 2000).

Recent pollution

The study of *A. b. tepida* through the Holocene sediments of Arketas suggests that dysaerobic conditions were not a one-off event, but were generalised during an interval of sub-unit 3.1. With the aim of evidencing anthropogenic effects on the recent evolution of this site, we have compared the occurrence of heavy metals Pb, As, with *A. b. tepida* morph C records in Arketas (Fig. 4). This graph represents the upper part of unit 1 and units 2 and 3 of the Arketas core. The age of these levels was estimated based upon the sedimentation rate obtained by decrease of isotope ^{210}Pb exc. (Pascual et al., 1998). The sedimentation rate estimate (0.96 cm yr $^{-1}$) for the lower estuarine area of Gernika, indicates an age of 1940 c.d. (calendar date) to the 50 cm core depth level and about 1970 c.d. to 20 cm core depth. The co-variant occurrence of As and *A. b. tepida* morph C is noticeable, and there is a very similar record of Pb (see Fig. 4). The maximum concentration of heavy metals (unit 3.1; 20 cm core depth) coincides with the maximum occurrence of *A. b. tepida* morph C. This indicates that distribution of *A. b. tepida* morph C is, at least partially, related to heavy metals. The same has been described by Sharifi et al. (1991) in Southampton (U.K.), where higher concentrations of Cu and Zn occurred in deformed specimens of *A. beccarii*. Comparable examples come from the mediterranean coast of Israel (Yanko et al., 1994) and from several sites in France and Brazil (Geslin et al., 1998), where deformities in specimens of *Ammonia* have been related to pollution from heavy metal contamination, changes in physical and chemical parameters, and/or shortage of nutrients. Nevertheless, the fact that we have found no deformed tests of this taxon in Arketas induces us to think that another factor, such as dysaerobia caused by eutrophization by nutrient inputs in several areas of this estuary (Orive et al., 1998), might have been responsible for at least part of the anomalous shift of *A. b. tepida* morph C in that time.

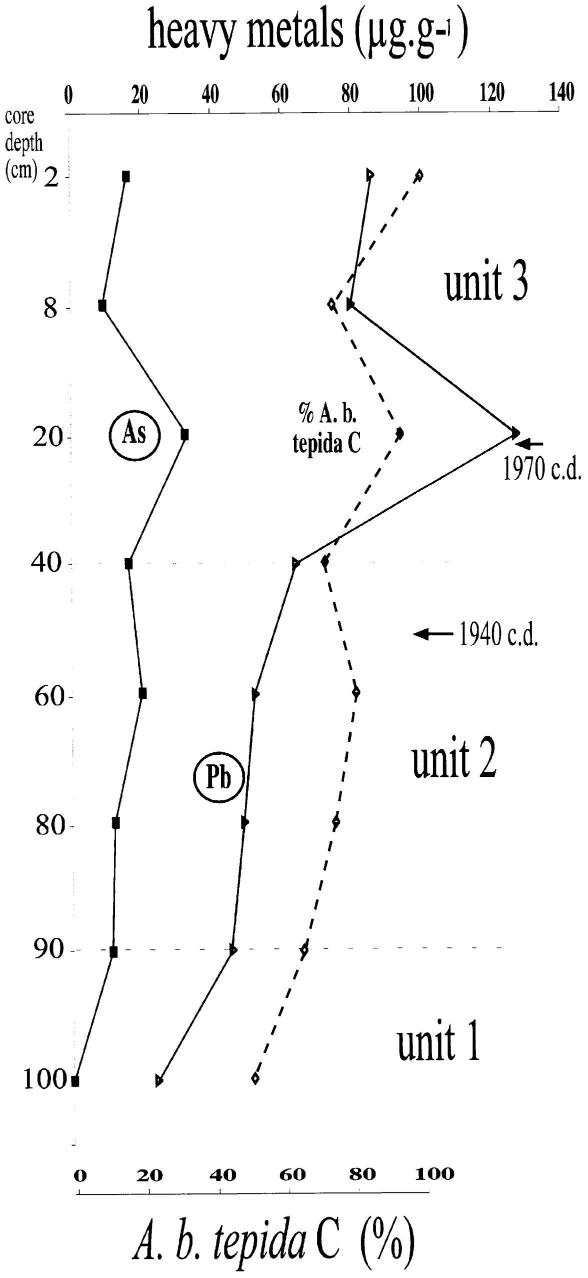

Figure 4. Distribution of heavy metals (As, Pb) and *A. beccarii tepida* morph C in Arketas sedimentary core.

Taking into account previous results, a secular evolution of the Arketas area can be proposed. During the final Holocene (up to 3500 years BP; unit 1) this site was a middle-outer estuarine settlement, euryhaline with low salinity. This episode ends with a marine transgressive pulsation. After an unrecorded, unknown time, recent sediments (unit 2) register a modern palaeogeographic position of Arketas which is located in a lower estuary environment. Anthropogenic activity by heavy metals is evidenced for the first time in this area at the limit between units 2 and 3 (around 1950 c.d.). In more recent times (unit 3.1.) an increase in anthropogenic activity is evident, with the maximum being reached in the 1970s, and a decrease from the 1980s. This recovery from pollution was probably a consequence of the creation of the Natural Reserve of the Biosphere of the Gernika estuary by UNESCO in 1984, which was the first conservationist legislation in this region.

Conclusions

Late Holocene evolution of an area of Gernika estuary (Southern Bay of Biscay) is evidenced by the study of calcareous microfaunas of foraminifers and ostracods, from a sedimentary core at Arketas pier. The occurrence of hypoxia and/or pollution events in the geological record is evidenced by the drastic reduction of microfaunal diversity indices as well as by the dominance of *A. b. tepida* morph C, a good biological indicator of stressful benthic estuarine conditions. The lower estuarine area of Gernika was submitted, about 3.8 ka to euryhaline conditions, with low-diversity microfaunal assemblages. A marine pulsation at about 3.5 ka was registered in this estuary by means of foraminifer and ostracod type-assemblages, the increase of *A. b. tepida* morph C, and the generalised increase of calcareous microfauna diversities. After an interval of lack of sedimentation, new marine influence is evidenced by the entrance of individuals living on the shelf and by planktonic foraminifers. In recent decades, a biotope similar to today's settled in this area, in the context of a stronger sedimentary filling of this part of the estuary.

In more recent times, an increase in anthropogenic activity is deduced based on the occurrence of heavy metals and biological proxies (*A. b. tepida*). The maximum of pollution being reached in the 1970s, and decreased from the 1980s, very likely as a consequence of the conservationist legislation to the Natural Reserve of the Biosphere of the Gernika estuary by UNESCO in 1984.

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Planktonic chlorophyll *a* and eutrophication in two Mediterranean littoral systems (Mallorca Island, Spain)

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Key words: chlorophyll *a*, eutrophy, estuary, harbour, western Mediterranean

Abstract

Two Mediterranean littoral systems of Mallorca Island have been studied using planktonic chlorophyll *a* (total concentration and seasonal variations) as trophic indicator. The studied systems are quite different in geomorphology, physicochemical trends and anthropic incidence. S'Albufera-Alcudia Bay is the drainage outlet of a coastal marsh, with a salt wedge estuarine regime. Andratx Harbour is a narrow bay affected by a high anthropic incidence due to the presence of a small fishing port and mainly a large leisure harbour. Both systems are affected by a typical Mediterranean climate and a negligible tide incidence. The present work is based on several year monitoring of physical, chemical and biological water parameters. In S'Albufera-Alcudia Bay, Chl *a* concentration ranged from 0.01 to 17.10 mg m^{-3}, the values dropping from the inland site to the sea. The phytoplankton biomass gradient takes its origin in physical and chemical differences. The variance is mainly related to the inland-sea water axis. Nitrogen and silica enriched inland waters are the main source of eutrophication, hence their relation with Chl *a*. These nutrients entering the Albufera are mainly derived from leaching from farmed soils. The transition from an inland eutrophic system to an oligotrophic marine one show the filter character of these coastal marshes. In Andratx Harbour, Chl *a* ranged from a non detectable concentration to 6.36 mg m^{-3}. The ultraplankton (<5 μm) was the most important contributor to total concentration. The ultraplankton in this system is related to influence of more oligotrophic open seawater. A greater proportion of microplankton (>20 μm) is related both to higher nutrient concentrations and a higher water column stability. In Andratx Harbour, the enclosure state seems very important for development of phytoplankton blooms.

Introduction

The increase of eutrophy in coastal systems has become more general during last decades (Larson et al., 1985; Vollenweider et al., 1992; Folch, 1999). Different sources are the causes of this eutrophication, mainly associated to human activities (Costanza & Greer, 1998). Although, our knowledge about the eutrophication processes in coastal waters is still limited at the moment. In a lot of cases, this process is compared with the eutrophication in limnological systems, mainly lakes and reservoirs which have been studied for long time. Recently, many studies focused on eutrophication processes and its forcing factors in coastal areas (Kennish, 1992; Jørgensen & Richardson, 1996a; Clark, 1998). The sources of eutrophication and their effects are different depending on the type of coastal systems (coastal marsh, estuaries, bays or large coastal systems). A recent increase of anthrophogenic use of littoral areas for residential and tourist activities is one of the most important causes of eutrophication. Tourism is specially important in economic development for small coastal countries and islands with limited economic options (Clark, 1998). In the Mediterranean Sea, the eutrophication phenomena have been mainly related to high populations and human use of the littoral (Ambio, 1979; Clark, 1992; Folch, 1999).

Two littoral systems of Mallorca Island have been studied in the present work. Mallorca is part of the Balearic Islands (Western Mediterranean Sea) and is affected by a typical Mediterranean climate (Colom, 1978). The Balearics are also characterised by very small tides, maximum amplitude about 20–30 cm (Alonso et al., 1990), so tide incidence is almost negligible. The studied systems are quite different in geomorphology, physicochemical trends and anthropic incidence.

One of the studied system, named S'Albufera-Alcudia Bay, is located in the NE of Mallorca Island and consists of the drainage outlet canal of a coastal marsh and the adjacent sea waters (Fig. 1). It features small-scale estuarine characteristics with a typical salt wedge regime (Puigserver et al., 2002). S'Albufera coastal marsh has a surface of about 24 km^2 and is separated from the sea by a sandbank 8 km long and 300–500 m wide. This wetland has been affected by human activities since second half of the nineteenth century, originally to drain it (Barceló & Mayol, 1980). Nowadays, it features an extensive network of drainage ditches which converge in a single outlet into the sea and some lagoons dispersed on the whole area (Martínez-Taberner & Mayol, 1995). Water replenishment occurs during torrential inputs as well as from semicontinuous intakes from springs and infiltration of wastewater and seawater (Martínez-Taberner et al., 1991). Waters are mainly holigohalines, with salinities about 0.5–5 psu (Martínez-Taberner, 1988). This wetland was declared a Natural Park in 1988 (Secona, 1990) and one of the aims for its rehabilitation was to maximise the water residence time. Since then, there has been an anthropic regulation of the outflow by means of diverse sluices. Although precise flux data are not available, annual outflow from S'Albufera to the sea caused by springs and infiltration has been estimated to be about 25–30 Hm^3 yr^{-1} (Jaume, 1980) and the park management sustains a gentle and continuos runoff through the year. Only during heavy rainfall periods, water discharges from temporary streams increase for short times the runoff flux, involving an annual mean extra-outflow of 23 Hm^3 yr^{-1} (Jaume, 1980). The adjacent sea waters are part of a large bay, Alcudia Bay, which is also affected by a large tourist resort on the north coast. The width on its mouth is near 14 km and its maximum depth about 35 m. Water salinity of 36.5–37.5 psu is highly regular for the whole bay, except for areas close to the Albufera outlet where salinities about 10–35.5 psu have been measured (Vives, 1992).

The second studied system, Andratx Harbour, is on the contrary a narrow and rocky bay located in the SW of Mallorca Island (Fig. 1). It is affected by a high anthropic incidence due to the presence of a small fishing port and mainly a large leisure harbour. It is also affected by a highly urban development on the whole littoral. The width on its mouth is near 1 km and its maximum depth about 25 m. There is any river discharge in this system or any other type of continuous inland water inputs. Only sporadic inland water discharges by means of temporary streams associated to rainfall regime are known, the main flows into the innermost site of the bay. No previous hydrological data are available from this system.

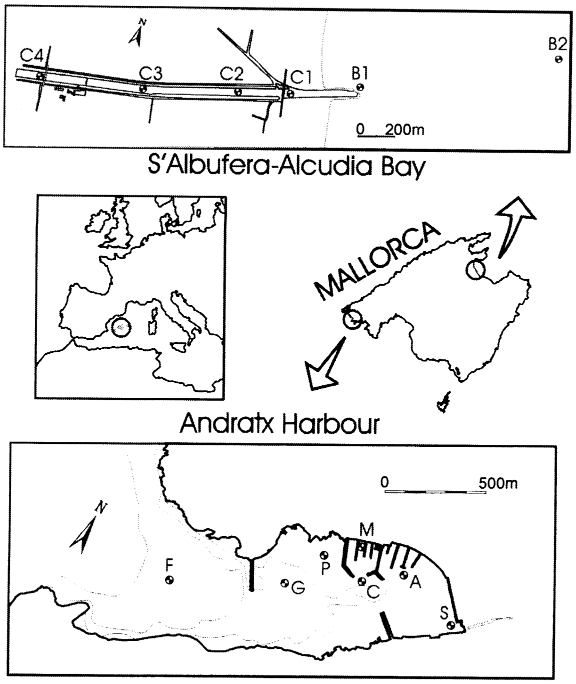


Figure 1. Location of S'Albufera-Alcudia Bay and Andratx Harbour on Mallorca Island (Western Mediterranean Sea). Sampling sites are indicated with black and white circles.

The aim of this work is to analyse the changes in chlorophyll *a* (Chl *a*), both in time and space, and to discuss them according to different eutrophication sources and different traits of the studied systems.

Materials and methods

At S'Albufera-Alcudia Bay, sampling was carried out near monthly during a complete year (December 1992–February 1994) bringing up to 14 sampling dates. Six sampling sites were selected (Fig. 1) and a whole of 157 samples were analysed. At Andratx Harbour, sampling was carried out seasonally from March 1995 to September 1998 over 3-month periods increasing up to weekly in some periods, bringing up to 25 sampling dates. Seven sampling sites were selected (Fig. 1) and a whole of 355 samples were analysed. In both cases samples were taken from just below the surface and about 20–25 cm above the bottom. Samples were taken using a peristaltic pump which enables precise sampling at different depths and not disturbing the sediment. At C4 site on the coastal marsh, only one sample was taken, due to water level seldom surpassed 20 cm. At the remaining sites into the canal water column was about 1 m. At the outermost site into Alcudia bay (B2) water level was about 13 m. At Andratx Harbour, water levels were from 2 to 4 m in the innermost area (S, A, M, C and P) and from 8 to 17 m in the outermost area (G and F). Additional samples from other levels were also taken when water column surpassed 5 m.

Salinity, temperature, oxygen, light intensity, turbidity and pH were determined *in situ* with specific sensors. Salinity and temperature were measured using a Microprocessor Conductivity meter WTW LF196. Oxygen concentration using a Microprocessor Oxymeter WTW OXI196. Turbidity using a HACH 2100P Turbidimeter. pH was measured using a WTW 196T pH-meter. Alkalinity was determined following Golterman et al. (1978) and Strickland & Parsons (1972). Nitrates, nitrites, silicates and phosphates were determined with a Technicon Autoanalyzer II following Grasshof (1969) for the first three and Tréguer & Le Corre (1975) for the last.

Pigment concentration was determined by filtration through Whatman GF/C, extraction in 90% acetone and measurement of absorbance with a Hitachi 2000 spectrophotometer. Chlorophyll *a* (Chl *a*) concentration was calculated according to Jeffrey & Humphrey (1975). In Andratx, 140 additional samples were taken from July 1996 to September 1998, 12 sampling dates, to analyse the pigment concentration of the size classes obtained filtering through 20 micra net, 5 micra net and GF/C filter.

Principal Components Analysis (PCA) have been used as statistical treatments for physical, chemical and biological data in order to obtain an integrated view. Seasonal mean Chl *a* concentration have been calculated using samples from March to May, June to August, September to November and December to February as spring, summer, autumn and winter, respectively, in order to study seasonal changes and to easier compare the two studied systems.

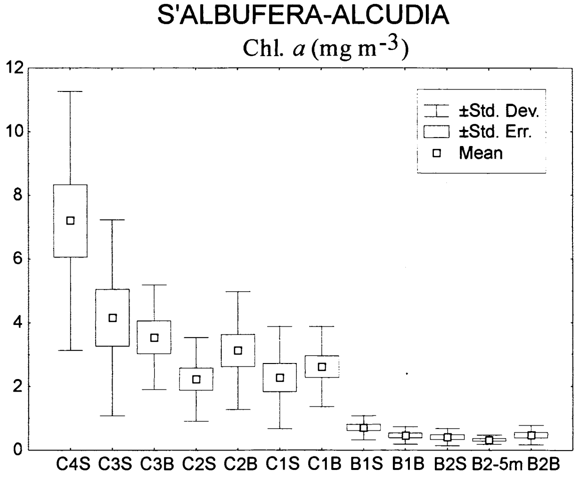


Figure 2. Mean chlorophyll *a* (Chl *a*) per sampling site in S'Albufera-Alcudia Bay (S: surface and B: bottom). Standard deviation and standard error are also shown.

Results

The main characteristic of S'Albufera-Alcudia Bay is the clear longitudinal gradients of physical and chemical parameters. Salinity ranged from 3.3 to 38.7 psu increasing to sea sites and in bottom levels (Table 1). In the coastal marsh, C4, salinity remained near 5 psu stating the brackish character of the Albufera waters. In the low canal, C1, salinity ranged from 5.5 to 31.1 psu at the surface, and from 5.9 to 36.9 psu at the bottom. Into the bay, B2 salinity ranged from 36.6 to 38.7 psu, no significant differences were observed from surface to bottom samples. A gradient of alkalinity was recorded on the opposite direction of salinity. Values ranged from 2.39 to 5.92 meq l^{-1}.

Temperature followed yearly solar cycles with winter minimum, 8.4 °C, and summer maximum, 31.7 °C. The change in temperature was higher in the coastal marsh, 21.4 °C, than into the bay, 14.8 °C. Which is caused by the shallower water column in the coastal marsh. Light intensity also followed yearly solar cycles and light limitation is not expected, values ranged from 13 to 752 μE m^{-2} s^{-1}.

Oxygen concentration in water was generally higher in the coastal marsh, values from 9.1 to 21.4 mg l^{-1} and 107 to 323% oxygen saturation. At the low canal values ranged from 3.2 to 15.6 mg l^{-1} and 50 to 138%. Values from 5.3 to 11.8 mg l^{-1} and 78 to 156% oxygen saturation were observed into the bay.

Nitrates, nitrites and silicates decreased from the Albufera towards Alcudia Bay (Table 1). Values were generally higher in surface than in bottom levels.

Table 1. Salinity, nutrient and chlorophyll *a* (Chl *a*) minima and maxima values per sampling site groups

		Sal. (psu)	Nitrate (μM)	Nitrate (μM)	Silicate (μM)	Phos. (μM)	Chl *a* (mg m^{-3})
S'Albufera–Alcudia							
C4		3.3–10.8	252–1164	0.42–27	57–220	0.03–0.13	2.69–17.10
C3/C2/C1	S	5.5–33.1	58–585	0.08–10	30–106	0.03–0.18	0.63–12.39
	B	5.9–36.9	7.8–224	0.03–3.6	6.78–74	0.03–0.37	0.57–5.82
B1/B2	S	17–38.7	0.05–261	0.01–1.61	0.47–68	0.03–0.41	0.01–1.89
	B	37–38.7	0.05–4.2	0.01–0.42	0.38–2.28	0.03–0.27	0.06–1.2
Andratx							
M/S	S	35.1–38.3	N.D.–5.38	0.01–0.26	0.7–17	0.02–0.19	0.25–4.71
	B	37–38.2	N.D.–1.08	0.01–0.13	0.9–3.29	0.01–0.15	0.21–6.36
A/C	S	37–38.2	N.D.–1.27	N.D.–0.12	0.73–2.92	0.01–0.17	0.25–2.80
	B	36.9–38.2	N.D.–1.15	N.D.–0.15	0.72–2.11	0.02–0.08	0.22–2.46
P/G/F	S	36.7–38.2	N.D.–1.05	N.D.–0.21	0.56–1.83	0.01–0.09	N.D.–1.57
	B	37–38.3	N.D.–0.57	N.D.–0.15	0.6–1.86	N.D.–0.05	0.03–2.41

Phosphates concentrations were low over the entire study. Maxima values up to 0.41 μM were found at the mouth of the canal and in the adjacent area into the bay. In canal sites C3, C2 and C1 bottom values were generally higher than surface ones.

Chlorophyll *a* (Chl *a*) concentration ranged from 0.01 to 17.10 mg m^{-3}, the values dropping from the Albufera to Alcudia Bay (Table 1). Annual mean values per sampling site and the ranges of variation also show the same decreasing gradient (Fig. 2). In this way, sampling sites have been gathered in three groups, which also agree with nutrients concentrations (Table 1). In the low canal, C2 and C1, mean Chl *a* values were higher in bottom level than in surface one (Fig. 2).

Monthly variation of Chl *a* shows significant peaks in the coastal marsh, C4, from February to April 1993 and from October to December 1993, those values were higher than 8 mg m^{-3} (Fig. 3). The change in time of Chl *a* is smaller at the low canal and specially into the bay. A winter or early spring peak can be identified in the low canal, C2 and C1 (Fig. 3). High values have been also found in summer and sporadically in autumn. In the low canal, C2 and C1, bottom values are generally higher than surface ones (Fig. 3). Into the bay Chl *a* maxima have been observed sporadically in summer and winter.

To clarify the seasonal pattern of variation, seasonal mean Chl *a* concentrations are shown in Figure 4. Seasonal means showed high values in spring and autumn in the coastal marsh, C4 (Fig. 4). As a general trend in the low canal, specially in bottom levels, and into the bay high mean Chl *a* has been found in summer. In some levels, high mean values have been found in other season, mainly in winter (Fig. 4).

Principal Component Analysis (PCA) was carried out using physical, chemical and biological monthly data (Fig. 5). Component 1 (43.6% of the variance) is highly determined by the salinity in one way and by nitrate, silicate, alkalinity, nitrite, oxygen and Chl *a* in the other way. This component is related with the inland-marine waters interaction and with the decrease of eutrophy. Component 2 (19.8% of the variance) is determined by alkalinity, phosphate and silicate in one way and pH and oxygen saturation in the other. Also with the position of the other parameters, Component 2 seems to be related to seasonal variations showing change from winter to summer. The latter agree with the change in time of phosphate concentration, which decreased from winter to summer due the increase of biological consumption.

In Andratx Harbour, no clear longitudinal gradients of physical nor chemical parameters were observed. Salinity values ranged from 35.1 to 38.3 psu (Table 1). Except for M site, values were equal to or higher than 36.7 psu and no significant differences were observed. Both temperature and light intensity followed solar annual cycle, with low values in late autumn, winter and early spring, and high values in summer. Temperature ranged from 14.3 °C to 28.5 °C. Differences in temperature have been observed between the inner sites and the outer ones. The inner-

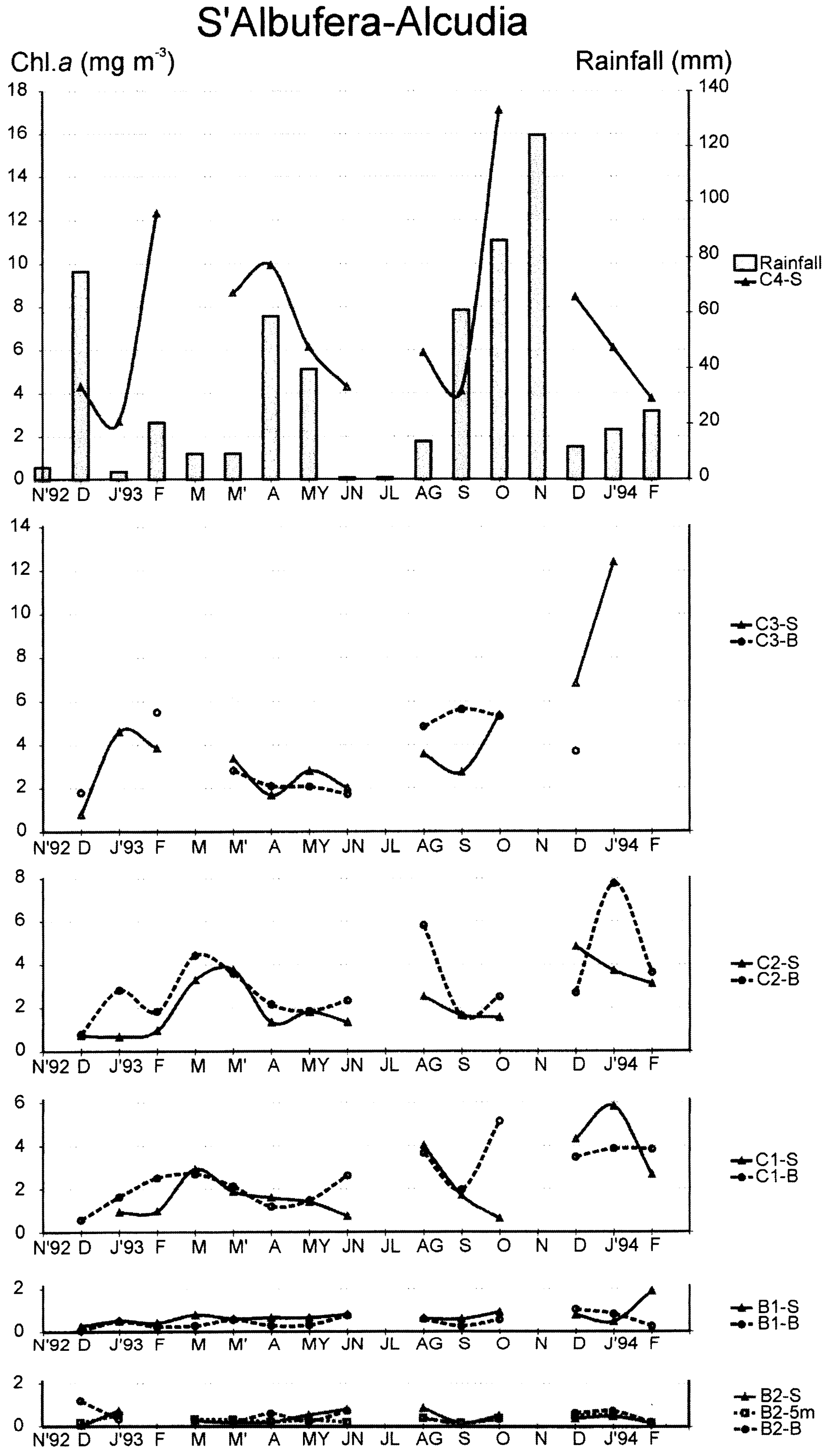
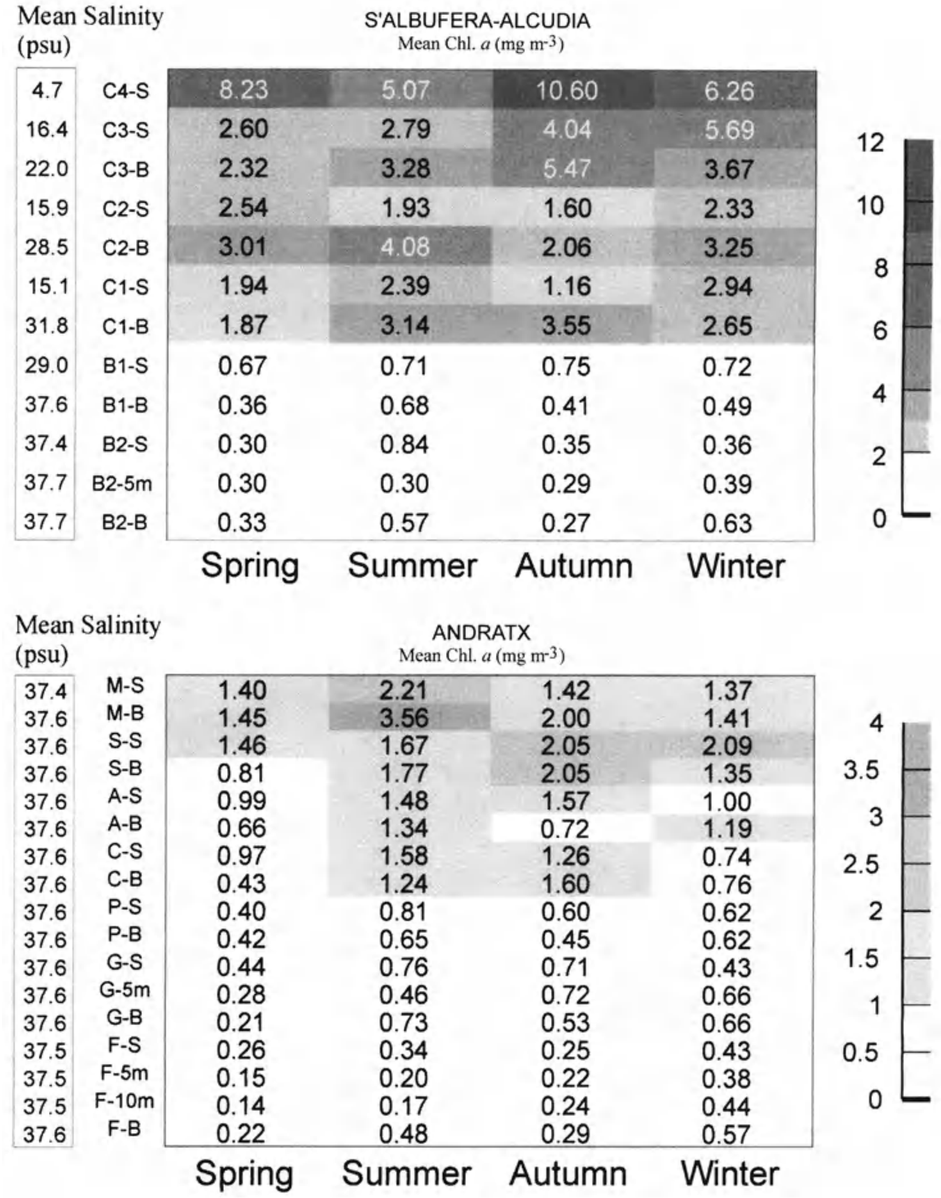


Figure 3. Monthly chlorophyll *a* (Chl *a*) per sampling site in S'Albufera-Alcudia Bay (S: surface and B: bottom). Rainfall regime during the monitoring is also shown.



S'ALBUFERA-ALCUDIA
Mean Chl. *a* (mg m-3)

Mean Salinity (psu)		Spring	Summer	Autumn	Winter
4.7	C4-S	8.23	5.07	10.60	6.26
16.4	C3-S	2.60	2.79	4.04	5.69
22.0	C3-B	2.32	3.28	5.47	3.67
15.9	C2-S	2.54	1.93	1.60	2.33
28.5	C2-B	3.01	4.08	2.06	3.25
15.1	C1-S	1.94	2.39	1.16	2.94
31.8	C1-B	1.87	3.14	3.55	2.65
29.0	B1-S	0.67	0.71	0.75	0.72
37.6	B1-B	0.36	0.68	0.41	0.49
37.4	B2-S	0.30	0.84	0.35	0.36
37.7	B2-5m	0.30	0.30	0.29	0.39
37.7	B2-B	0.33	0.57	0.27	0.63

ANDRATX
Mean Chl. *a* (mg m-3)

Mean Salinity (psu)		Spring	Summer	Autumn	Winter
37.4	M-S	1.40	2.21	1.42	1.37
37.6	M-B	1.45	3.56	2.00	1.41
37.6	S-S	1.46	1.67	2.05	2.09
37.6	S-B	0.81	1.77	2.05	1.35
37.6	A-S	0.99	1.48	1.57	1.00
37.6	A-B	0.66	1.34	0.72	1.19
37.6	C-S	0.97	1.58	1.26	0.74
37.6	C-B	0.43	1.24	1.60	0.76
37.6	P-S	0.40	0.81	0.60	0.62
37.6	P-B	0.42	0.65	0.45	0.62
37.6	G-S	0.44	0.76	0.71	0.43
37.6	G-5m	0.28	0.46	0.72	0.66
37.6	G-B	0.21	0.73	0.53	0.66
37.5	F-S	0.26	0.34	0.25	0.43
37.5	F-5m	0.15	0.20	0.22	0.38
37.5	F-10m	0.14	0.17	0.24	0.44
37.6	F-B	0.22	0.48	0.29	0.57

Figure 4. Mean seasonal chlorophyll *a* (Chl *a*) per sampling site (S: surface and B: bottom) in both studied systems. Annual mean salinity is also shown.

most area of the harbour is subject to higher warming and cooling than the outer area. Light intensity values ranged from 13 to 815 μE m^{-2} s^{-1}.

Turbidity ranged from 0.2 to 1.56 NTU at the outermost site, F, and from 1.05 to 11 NTU at the more enclosed site, M. Mean turbidity in M (3.82 NTU) and S (2.68 NTU) were the highest. A and C showed the same mean turbidity (1.56 NTU). And P, G and F showed lower turbidity means (1.04, 0.7 and 0.57 NTU).

Oxygen concentration in water ranged from 3.7 to 10.7 mg l^{-1} and oxygen saturation from 46 to 149%. No significant differences have been observed in space and an annual pattern of variation is not clear.

Nutrient concentrations were low over the entire study area and most part of the year. Nutrient concentrations ranged from non detectable values (N.D.) to 5.38 μM of nitrate, 0.26 μM of nitrite and 0.19 μM of phosphate. Silicate ranged from 0.56 to 17 μM. A yearly cycle can not be readily observed. Based on

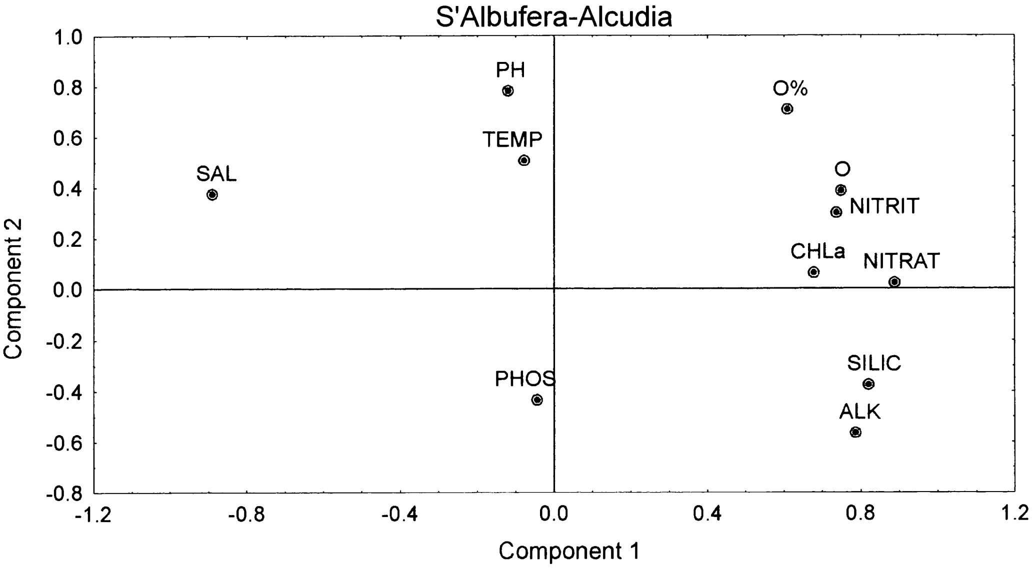


Figure 5. Position of physical, chemical and biological parameters of S'Albufera-Alcudia Bay in the space defined by Principal Components 1 and 2.

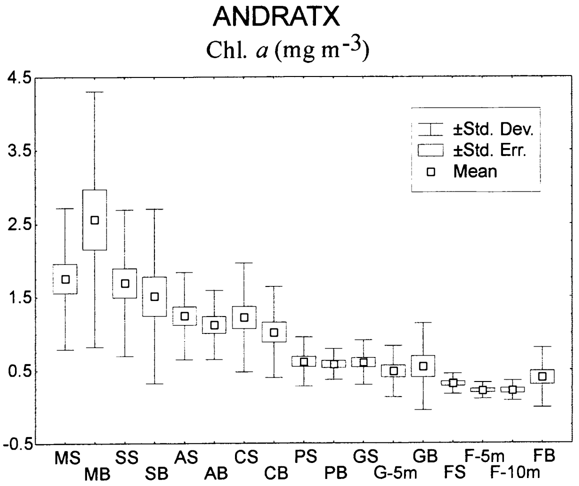


Figure 6. Mean chlorophyll *a* (Chl *a*) per sampling site in Andratx Harbour (S: surface and B: bottom). Standard deviation and standard error are also shown.

nutrient concentrations the sampling sites have been assembled in three groups (Table 1). In general, S and M showed the highest nutrient concentration; P, G and F showed the lowest nutrient concentration; A and C go between the other groups. These assemblages agree with turbidity data showed above. In general, values were slightly higher in surface waters (Table 1).

Chl *a* ranged from a non detectable (N.D.) concentration to 6.36 mg m^{-3} (Table 1). Chl *a* also agree with the site assemblages based on nutrient concentrations. M and S sites showed higher values and P, G and F sites showed lower values. Annual mean values per sampling site and the ranges of variation also agree with the above groups (Fig. 6). The highest range of variation has been found in the inner harbour and the smallest in the outermost site.

Seasonal mean Chl *a* concentration are shown in Figure 4. High mean Chl *a* concentrations were found mainly in summer and autumn. In some cases, high values have been also found in winter. Low mean Chl *a* concentrations have been found mainly in spring (Fig. 4). Concerning the distribution of Chl *a* per size classes, Chl *a* related to ultraplankton ($<5\ \mu$m) was the most important contributor to total concentration in all sampling sites (Fig. 7). This size fraction contributed from 39% (bottom S sample in winter) to 85% (surface G sample in spring) of total concentration. Chl *a* related to microplankton ($>20\ \mu$m) was the second fraction in abundance, ranging from 8% (surface G sample in spring) to 50% (bottom S sample in winter) of total concentration. Finally, Chl *a* related to large nanoplankton (20–5 μm) contributed from 7% (surface G sample in spring) to 23% (bottom M sample in autumn) of total concentration. So large nanoplankton showed the smallest range of variation (Fig. 7). As a general trend, low ultraplankton percentages were found in the innermost area of the harbour (M, S and A sites), where microplankton expressed a significant proportion main part of the year (Fig. 7). On the con-

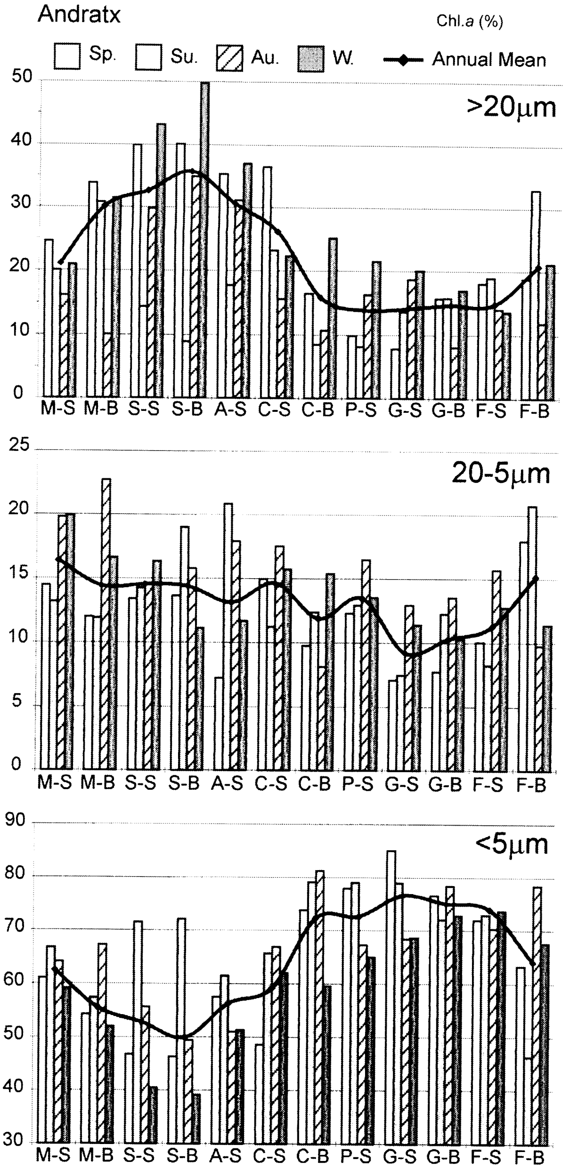


Figure 7. Annual and seasonal mean size fraction contribution to total chlorophyll *a* (Chl *a*) in Andratx Harbour. (Sp.= spring; Su.= summer; Au.= autumn; W.=winter; S: surface and B: bottom.)

trary, high ultraplankton percentages were found in the outermost area (P, G and F). Seasonal mean variation showed high contributions of microplankton mainly in winter and spring; high contributions of ultraplankton mainly in summer and autumn; and high contributions of large nanoplankton mainly in autumn and also in winter or summer depending on sampling site (Fig. 7). No significant correlations have been found between each size fraction and single hydrological parameters.

As in S'Albufera-Alcudia, a Principal Component Analysis (PCA) was carried out using physical, chemical and biological single data from Andratx Harbour (Fig. 8). Component 1 (23% of the variance) is highly determined by nutrients (nitrate, silicate, phosphate) and alkalinity in one way, and by ultraplankton on the opposite one. This component seems to be related with the trophic state. Component 2 (18.2% of the variance) is highly determined by Chl *a* size fractions (Fig. 8). The position of other parameters such turbidity and oxygen also agree with that Component 2 seems to be related with water column stability. The low variance that explain these two components shows the significance of a lot of factors interaction in the system.

As a general trait, Andratx Harbour trophic state is lower than that of S'Albufera, both the coastal marsh and most of other canal sites (Figs 2 and 6). Throphic state of Alcudia Bay is similar to that of the outermost area of Andratx Harbour and lower than that of the inner harbour.

Discussion

S'Albufera-Alcudia Bay showed a phytoplankton gradient caused by physical and chemical heterogeneity. It features common characteristics of an estuarine system. High silicates and very high nitrates concentrations have been observed in the coastal marsh, S'Albufera, decreasing to the sea, Alcudia Bay. Higher concentrations found in surface layer also suggest their relation with freshwater inputs. These values, specially those of nitrates, were significantly higher than those of other Mediterranean estuaries as low river Rhône (Lochet & Leveau, 1990), and those of other world-wide common estuarine ecosystems (Boynton et al., 1982). On the contrary, phosphate concentrations were low compared with other estuarine systems (Boynton et al., 1982) and even similar to those of surface Mediterranean Sea (Estrada et al., 1985). Waters from the Albufera are rich in nitrogen compounds due to rural inputs in the upper areas. This is the main source of eutrophication in the system, hence their relation with Chl *a*. These nutrient entering the Albufera are mainly derived from leaching from farmed soils in the limits of the coastal marsh (Martínez-Taberner et al., 1990). An increase of nitrate concentration has been observed with regard to values of Martínez-Taberner et al. (1990), which suggests an increase of farming fertilising or a reduction of the

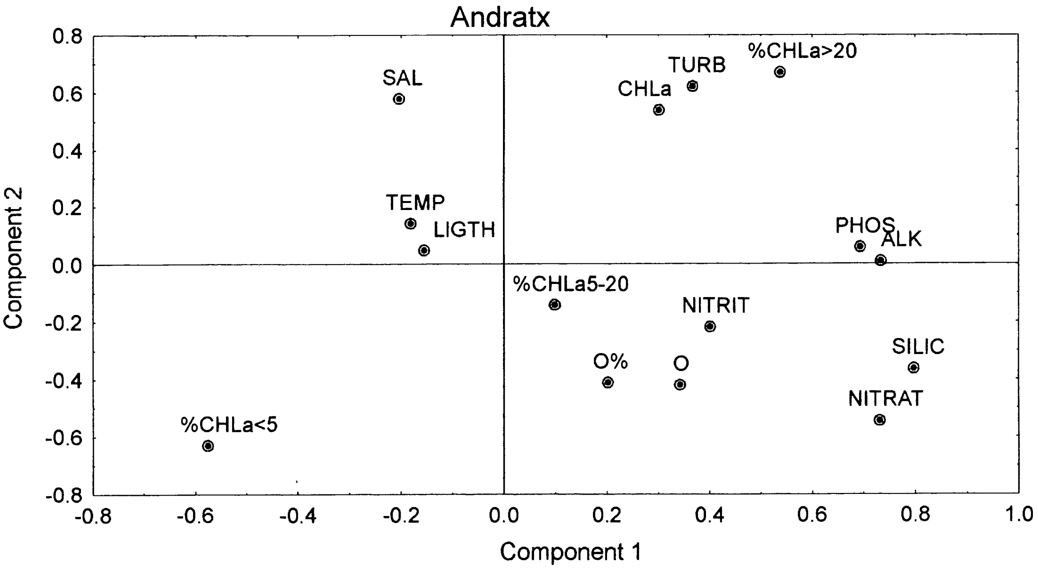


Figure 8. Position of physical, chemical and biological parameters of Andratx Harbour in the space defined by Principal Components 1 and 2.

nutrient trap character of the coastal marsh. Phosphate concentration in the lower part of the canal has experienced a heavy decrease which can be associated to the introduction of new sewage farms in the adjacent area. Similar trends of nutrients changes, increase of nitrate and decrease of phosphate, have been observed in Alcudia Bay compared with previous data (Vives, 1992).

In this system, the change of physical, chemical and biological parameters is mainly related to inland–sea water interaction. Which depends mainly on natural factors, as rainfall pattern, but also on anthropogenic regulation of water fluxes in the canal.

Chl *a* concentrations in S'Albufera and in the canal are comparable to values found in larger estuarine systems (Boyton et al., 1982). On the other hand, values of Alcudia Bay are most part of the year similar to those of open Mediterranean waters (Innamorati et al., 1985; Estrada et al., 1985; Martínez et al., 1990), and similar to those of other Mediterranean coastal waters the rest of the year (Masó & Duarte, 1989; Riaux–Gobin et al., 1998).

Chl *a* peaks in the coastal marsh seem to be related mainly with the rainfall regime in the area (Fig. 3). Nutrient inputs by runoff in spring and autumn are the main cause of phytoplankton growth in the inner marsh, when temperature is not a limiting factor. The latter follows the model of Dugdale & Goering (1967) for temperate estuaries. Freshwater incidence on phytoplankton community has been related to growth of Clorophyceae (Puigserver et al., 2002). The effect of nutrient rich inland waters on the low canal and into the bay is small, so Chl *a* ranges of variation are low. Summer Chl *a* maxima coincides with peaks of phytoplankton cell abundance, with growth of Cyanophyceae and Cryptophyceae in the low canal (Puigserver et al., 2002) and of Dinophyceae and Bacillariophyceae into the bay (Moyà et al., 1995). These phytoplankton blooms at the low canal and into the bay should be related with breakdown of organic matter and recycling of phosphorus from the sediment. In this case, phytoplankton development follows the model described by Kemp & Boynton (1984) also for temperate estuaries. Nutrients inputs by other causes such macrophytes senescence, mainly in autumn (Martínez-Taberner et al., 1990), or sporadic increases of sewage discharges are likely in this system. So high Chl *a* values found in other periods could be the result of some of these eutrophication processes.

The gradient from an eutrophic system, S'Albufera, to an oligothrophic one, Alcudia Bay, and their different phytoplankton biomass annual pattern show the filter character of these type of coastal systems. The water movement caused by runoff tends to favour initials stages of phytoplankton succession and agrees with high contribution of small nanoflagellates (mainly <5 μm) found in the canal (Puigserver et al., 2002). This flux also favour the marine dispersion of nutrients coming from the coastal marsh.

Andratx Harbour represents a different case. No significant differences have been observed in salinity, light nor oxygen in water. Temperature showed low spatial heterogeneity caused by the greater warming and cooling of water in the innermost area, which is related to the fewer depth and higher water stability.

Nutrient concentration was low over the entire study. Values were similar to those of oligotrophic Mediterranean waters (Estrada et al., 1985; Lochet & Leveau, 1990). Based on turbidity, nutrient concentration and Chl *a* three areas with different trophic state have been distinguished in the harbour. Higher values have been found in the most enclosed sites. The eutrophication sources in this system are nutrient inputs mainly derived from urban sewage on the whole shore line and also from temporary streams in the inner harbour. Spatial heterogeneity does not seem associated to any clear longitudinal gradient.

Although nutrient concentration suggest an oligotrophic character, Chl *a* concentration suggest a mesotrophic character of the inner harbour. Values were similar to those observed in other coastal waters (Innamorati et al., 1985; Videau & Leveau, 1990) and harbour systems (Rodríguez & Vives, 1984). In the outermost area, values were similar to those of open Mediterranean Sea (Estrada et al., 1985; Raimbault et al., 1988).

Chl *a* related to ultraplankton (<5 μm) has been the most important contributor to total phytoplankton biomass in the whole system. The greater contribution of ultraplankton is related with more oligotrophic open seawaters. Contribution of Chl *a* related to large nanoplankon (20–5 μm) has been more or less constant. Nanoplankton of different taxonomic classes, mainly Cryptophyceae, Prasinophyceae and Prymnesiophyceae among others, is the most abundant phytoplankton in this system (Puigserver, unpublished data). The increase of organic matter breakdown due to high temperatures and the rise of sewage discharges related to increase of population and movement of vessels explain the summer phytoplankton development. The last with high contribution of small sized fractions. Nutrient inputs related to autumn rainfall, which involve 219 mm and 38% of annual amount (Guijarro, 1986), support high phytoplankton biomass of the small size fractions. The small phytoplankton would be favoured by a turbulent environment (Margalef, 1978).

On the contrary, the greater contribution of microplankton (>20 μm) is related to more eutrophic waters linked to high water stability. Winter Chl *a* maxima in the inner harbour (mainly in S sampling site) have been related to high contribution of microplankton and development of *Alexandrium minutum* Halim red tides (Moyà et al., 2000). The larger algae have low growth rates and need some time to develop (Sournia, 1981). The nuisance phytoplankton blooms can be among others one of the secondary effects that alert of an eutrophication process, although cultural eutrophication is not always implicated in bloom formation (Jørgensen & Richardson, 1996b). Causes of harmful algal blooms are not well known. They have been related to human activities, to introduction of exotic species in ship ballast, to nutrient enrichment and to failure of controls by grazers (Valiela, 1995). Winter rainfall of about 180 mm has been measured in Andratx and involves 31% of annual total (Guijarro, 1986), so it has to be considered an important source of nutrients to the bay. Most part falls in early winter as a sequel of the rainy season. Although water temperature measured in late winter remained about 14.5–15.2 °C, periods of high stability following the storms season can induce phytoplankton blooms. This mechanism is considered to be similar to that described by Zingone et al. (1995) to explain autumn phytoplankton blooms in the Gulf of Naples.

Mainly the phytoplankton heterogeneity and the low nutrient concentration suggest that the enclosure state is very important for phytoplankton development in this system. The enclosure rends difficult the transport of nutrients to the open sea and favours the development of massive coastal phytoplankton blooms. Water stability enhances phytoplankton growth, which uses the nutrients incoming to the system. In this way, nutrient concentration in water amount remain low and the study of biotic indicators become necessary to determine the trophic state of the system.

The studied systems showed significant differences in throphic state. The causes are mainly related to different eutrophication sources, both the nutrient inputs and the hydrodynamics of the system. Despite nutrient discharges from S'Albufera to the sea, the outflow flux and the hydrography of a large bay favours the marine dispersion. On the contrary, in Andratx Harbour the enclosure created by breakwaters and successive mooring quays in an area without tide incidence minimises the flow mixing effect and nutrient marine dispersion. The enclosure appeared to be very important for phytoplankton outburst development.

Acknowledgements

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Physiological response of *Spisula subtruncata* (da Costa, 1778) to different seston quantity and quality

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Key words: bivalve, feeding behaviour, filtration, ingestion, regulation

Abstract

Individuals of the bivalve *Spisula subtruncata* were fed a mixed diet comprising of sea water enriched with the diatom *Phaeodactylum tricornutum* and ashed silt within a range of concentrations, simulating natural conditions above pseudofaeces threshold. The designed ranges for total particulate matter were between 10 and 30 mg l^{-1} and organic content of seston 15–40%. Filtration rate, rejection rate, ingestion rate and absorption rate were measured at those different conditions. Filtration rate and rejection rate were significantly correlated to total particulate matter and percentage of organic matter, with higher rates at higher values of total particulate matter and lower values of percentage organic matter. Ingestion rate was maintained at similar levels in all the treatments and organic enrichment of the ingested food occurred due to preingestive selection of the filtered material. A differential absorption rate occurred at different levels of organic matter in the diet with high rates at high values of the organic content of the diet. *S. subtruncata* showed different physiological responses to changes of the food conditions: (1) Increase of pseudofaeces production at increasing levels of particulate matter, (2) preingestive selection of organic material which enriched the organic fraction of ingested food, (3) stabilized ingestion rate and (4) increase of the absorption rate at high organic levels of the seston.

Introduction

Spisula subtruncata (da Costa, 1778) is a common bivalve which lives in coastal areas of Europe with a distribution from Norway to the Mediterranean and the Atlantic coast of Morocco (Tebble, 1966). The habitat of this species in the North Sea (sandy bottoms between 2 and 30 m) represents an environment where the concentration of suspended particulate matter (seston) may be variable in time as a result of resuspension of fine sediments during periods of high current velocity, wind-wave activity and storm events, especially in the shallow bottoms. For example, in the Dutch coastal area, seston concentrations in the water column can vary between 2 and 150 mg l^{-1}, with values between 5 and 10 mg l^{-1} being most frequent (RIKZ, 1999). Between 5 and 40% of this seston may be organic in nature, with Chlorophyll concentrations between 0 and 11 μg l^{-1}, depending on weather conditions, season or location within the coast. The effects of the variability of suspended particles in the water column has never been studied up to date in *S. subtruncata*, but it has been described for other bivalve species both in temperate (Bayne et al., 1989; Iglesias et al., 1996; Navarro & Widdows, 1997) and tropical waters (Yukihira et al., 1999; Wong & Cheung, 1999).

There is a controversy about the effects of the quantity and quality of seston in the feeding processes of bivalves. Jørgensen (1990, 1996) have suggested that responses to environmental changes are determined solely by the physical properties of the cilliary mechanisms of pumping and filtration. On the other hand, several authors (Bayne et al., 1989, 1993; Iglesias et al., 1992, 1998; Navarro et al., 1996; Navarro & Widdows, 1997; Hawkins et al., 1998; Yukihira et al., 1999; Wong & Cheung, 1999; Pouvreau et al., 2000) have observed bivalve filter feeding resulting in relative constancy of rates of absorption and ingestion.

Table 1. Number (*N*), shell length (SL: mm) and ash free dry weight (AFDW: mg) of the animals used in the different experiments (Exp) at different levels of total particulate matter (TPM: mg.l^{-1}) and percentage of organic matter (%OM). Date (year/month/day). Mean value ± standard deviation

Exp	Date	TPM	%OM	*N*	SL	AFDW
1	990804	12.59 ± 0.67	16.21 ± 1.33	7	25.4 ± 1.5	130.0 ± 19.7
2	990805	11.49 ± 0.41	26.41 ± 2.05	7	25.3 ± 1.1	118.6 ± 15.4
3	990802	10.66 ± 1.76	35.71 ± 1.65	7	25.3 ± 1.4	114.3 ± 16.0
4	990720	22.44 ± 0.95	17.53 ± 2.07	7	24.5 ± 1.2	103.6 ± 9.8
5	990723	18.98 ± 0.08	25.13 ± 0.63	7	25.3 ± 1.3	113.2 ± 21.2
6	990730	21.52 ± 0.56	43.66 ± 4.18	7	25.6 ± 1.6	119.4 ± 45.8
7	990719	29.48 ± 2.64	14.34 ± 0.99	7	24.8 ± 1.5	92.8 ± 22.5
8	990803	30.28 ± 0.8	25.17 ± 1.72	7	25.4 ± 1.5	132.0 ± 17.3
9	990729	30.31 ± 2.57	39.17 ± 1.41	7	24.7 ± 0.8	116.9 ± 33.5

The following modes of regulating the ingestion rate of food have been described: (1) changing the pumping or the clearance rate (Navarro et al., 1992, 1994; Iglesias et al., 1996; Navarro & Widdows, 1997), or (2) increasing rejection of pseudofaeces above a certain threshold of seston quantity and quality (Iglesias et al., 1992; Navarro et al., 1992; Navarro & Widdows, 1997). There are other mechanisms to increase the food ingestion and absorption by a preingestive selection of organic matter from filtered material (Bayne et al., 1993; Navarro & Iglesias, 1993; Iglesias et al., 1996; Hawkins et al., 1998; Wong & Cheung, 1999) and adjustment of the absorption efficiency to changes in gut passage time (Bayne et al., 1989; Navarro et al., 1994) or the gut content (Hawkins et al., 1990). Little information about feeding and ecophysiology of *Spisula subtruncata* is available (Møhlenberg & Riisgård, 1979; Kiørboe & Møhlenberg, 1981; Møhlenberg & Kiørboe, 1981). Research related to its growth in the North sea has been carried out recently (Degraer, 1999).

The aim of the experiment reported here is to determine how physiological rates are affected in *Spisula subtruncata* by different diets. The hypothesis is that this filter feeder is able to respond to variation in food quality and quantity by adjustments of physiological rates.

Material and methods

Collection and maintenance

Individuals of *Spisula subtruncata* were dredged from populations in the Molengat (North of The Netherlands) during June and July 1999 and transported to the Field station of RIKZ at Jacobahaven (Oosterschelde, S.W. Netherlands) where the experiments were executed. Selected individuals for experiments had a shell length of around 25 mm (Table 1). Acclimatisation to laboratory conditions proceed for one week. During this time the animals were placed in a container with fine sand and natural sea water pumped continuously from the coast. Parameters from the water were similar to those from the place of collection of the animals. Afterwards 7 individuals of *S. subtruncata* were placed in experimental chambers and fed on experimental diets for 5 h prior to start of the measurements. Collection of biodeposits were carried out 8 h after the start of the experiment. Two samples of biodeposits were collected for each individual on each experiment.

Experimental diets

Dietary composition included the following components: (1) Cells of the pelagic diatom *Phaeodactylum tricornutum* and (2) silt particles (<50 μm) previously ashed for removing the organic material. The mixtures of algaes and silt were added to natural sea water diluted with filtered sea water in a reservoir tank. A mixer was placed in the bottom and suspension of particles was promoted by aeration. A pump provided a flow of water through each chamber (2.8 l h^{-1}) in which the depletion of particles by the clams was below 30% of the total. For making and controlling the diets, the concentration of particles was monitored with a Coulter Counter fitted with a 100 μm tube.

The designed ranges for the diets were: total particulate matter (TPM: mg l^{-1}) 10–30 mg l^{-1} and percentage organic matter of seston 15–40%. Details of the different experimental diets are shown in Table 1. In each experiment, 3 water samples were taken during the experimental period from the inflow of the control chambers. For determinations of TPM, water samples (1 l) were filtered onto pre-ashed (450 °C for 4 h) and weighed GFC filters, rinsed with sea water-isotonic ammonium formate and dried at 80 °C. The dry weight of retained material gave the TPM and the weight loss on ignition at 450 °C for 4 h gave the ash weight or particulate inorganic matter (PIM: mg l^{-1}) from which the particulate organic matter (POM: mg l^{-1}) was derived (POM = TPM − PIM). The percentage organic content of suspended matter was computed as: % OM = (POM × 100)/TPM.

Collection of biodeposits and calculations of rates

During the experiment, faeces and pseudofaeces were collected separately at time intervals of 2–3 hours by using a Pasteur pipette. Biodeposits were separately filtered onto preashed and preweighed GFC filters. The total, inorganic and organic mass of biodeposits were determined by methods similar to those described above for sea water samples. The following physiological rates were calculated: Filtration rate (FR: mg h^{-1}), rejection rates of pseudofaeces (RR: mg h^{-1}), ingestion rate (IR: mg h^{-1}) and absorption rate (AR: mg h^{-1}). Referring to Conover (1966), absorption of inorganic matter through the digestive system is negligible, in this case the sum of inorganic rejection rate of pseudofaeces and inorganic egestion rate of faeces was considered to represent the inorganic filtration rate (IFR: mg h^{-1}), hence, clearance rates (CR: l h^{-1}) were estimated as CR = IFR/PIM. Filtration rate (FR: mg h^{-1}) was calculated as FR = CR × TPM and filtration rate of particulate organic matter (OFR: mg h^{-1}) as OFR = CR × POM. Ingestion rate was calculated as IR = FR − RR and the organic ingestion rate (OIR: mg h^{-1}) as the difference between OFR and the organic rejection rate (ORR: mg h^{-1}). The rate of food absorption (AR: mg h^{-1}) was computed as the difference of the organic rate of ingestion and the organic rate of egestion of faeces. Preingestive selection efficiency (SE) of the filtered organic matter was estimated as SE = 1 − (p/f), where p is the organic content of the pseudofaeces (p= ORR/RR) and f is, as defined above, the organic content of suspended matter (f= POM/TPM). When SE = 0, there is no selection (f=p) and when SE = 1, there is a complete selection and ingestion of organic particles.

Size standardization of physiological rates

Once the physiological measurements were completed, shell length of each individual was recorded to the nearest 0.1 mm as well as the ash-free dry weight of the soft tissues (weight after dry the soft tissues at 80 °C during 24 h − weight after calcination at 520 °C during 4 h). Rates were assumed to scale with body size, and consequently, measurements were standardised to an equivalent of 150 mg ash free dry tissue of *S. subtruncata* by using the formula $Y_s = Y_e * (150/W_e)^b$, where Y_S = physiological rate of a standard-sized animal, Y_e = uncorrected physiological rate, W_e = ash-free dry weight (mg) of the experimental animal and b = the weight power established for clearance rate of the bivalve filter feeders (b=0.58; Bayne & Newell, 1983), which has been assumed to represent the size dependence for rates of food processing in general.

Statistical analysis

The effects of different seston concentrations and percentage organic matter values on the physiological responses of *Spisula subtruncata* was tested with Two-way analyses of variance (ANOVA). The significance of differences between treatments was analysed with a Tukey–Kramer procedure as post-hoc test (Sokal & Rohlf, 1995). Regression analysis were performed with Systat 9.0. A significance level of 5% was used in all tests.

Results

Filtration rate

Total filtration rate (FR: mg h^{-1}) was tested at different levels of organic matter content (%OM) and total particulate matter (TPM: mg l^{-1}) of the diet (Table 2A). FR was significantly higher at high TPM and at low percentages of organic matter (Fig. 1).

Rejection rate & preingestive selection of the organic material

Rejection rate of pseudofaeces (RR: mg h^{-1}) increased significantly with FR and the regression equation is RR = 0.78 FR −1.34 (R^2=0.85, N=116, p<0.01). RR reached higher values at increasing TPM

Table 2. Statistical test of influences of total particulate matter (TPM) and percentage organic matter (% OM) on filtration rate (A), Rejection rate (B), Selection efficiency (C) and Absorption rate (D) of *Spisula subtruncata* in the different experimental conditions. Values are degrees of freedom (df), mean square (MS) and probability (*P*) of ANOVA

Source of variation	df	MS	*P*
(A) Filtration rate			
TPM	2	113.29	<0.001
%OM	2	109.90	<0.001
TPM x %OM	4	4.06	0.126
Error	107	2.20	
(B) Rejection rate			
TPM	2	62.10	<0.001
%OM	2	94.74	<0.001
TPM x %OM	4	3.69	0.071
Error	107	1.18	
(C) Selection efficiency			
TPM	2	0.003	0.853
%OM	2	22.08	<0.001
TPM x %OM	4	0.30	0.878
Error	103	0.02	
(D) Absorption rate			
TPM	2	0.121	0.101
%OM	2	1.133	<0.001
TPM x %OM	4	0.033	0.112
Error	101	0.017	

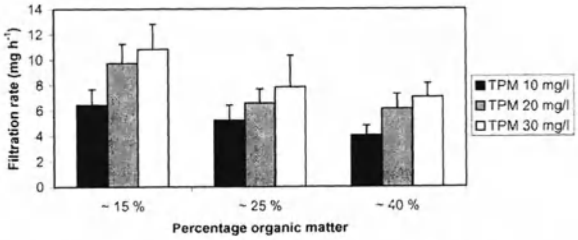

Figure 1. Filtration rate (mg h^{-1}) of *Spisula subtruncata* as a response to different concentrations of total particulate matter (TPM: mg l^{-1}) and at different levels of organic matter. Mean value + standard deviation.

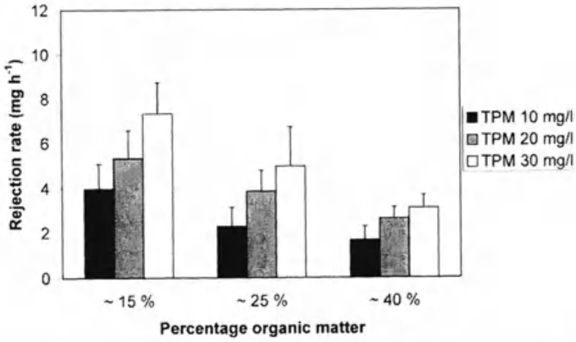

Figure 2. Rejection rate (mg h^{-1}) of *Spisula subtruncata* as a response to different concentrations of total particulate matter (TPM: mg l^{-1}) and at different levels of organic matter. Mean + standard deviation.

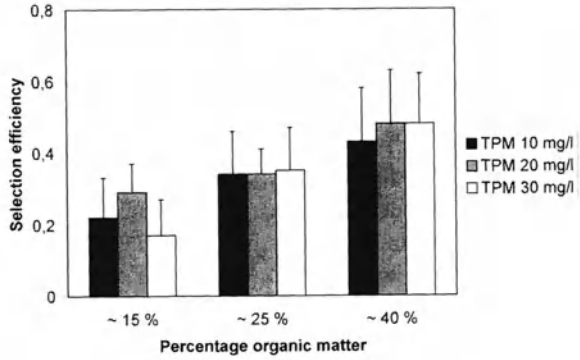

Figure 3. Preingestive selection efficiency (%) at different total particulate matter concentrations (TPM: mg l^{-1}) and percentages of the organic matter from the diet. Mean value + standard deviation.

and decreased at high percentages of organic matter within the same level of seston concentration (Fig. 2). The different treatments resulted in significantly different rejection rates of *S. subtruncata* at different seston quantity and quality (Table 2B). Together with the rejection of pseudofaeces preingestive selection of organic material occurred with a certain selection efficiency (SE). SE is significantly correlated with the quality of seston but not with the quantity (Table 2C). SE reaches high values at high percentages of organic matter in the diet (Fig. 3).

Ingestion rate & absorption rate

Ingestion rate showed no significant relation with different levels of seston concentration and percentage organic matter from the diet (Fig. 4). The organic ingestion rate (OIR: mg h^{-1}) was a function of SE with the following equation: OIR = 2.59 SE + 0.28 (R^2=0.41; N=111; p<0.01), both physiological parameters are dependent on the organic matter content as well. As described above, SE was high at high percentages of organic matter in the diet and then OIR is increased as a function of SE. However, values of OIR are underestimated by this procedure because we could not quantify the mucus rejection rate as a component of the organic rejection rate.

A positive relation occurs between the absorption rate of the ingested food (AR: mg h^{-1}) and the percentage organic matter (% OM:%) of the diet but

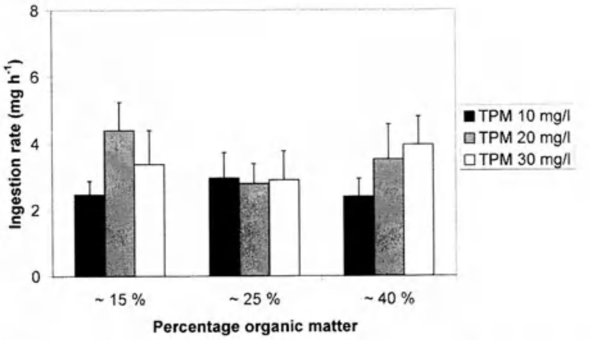

Figure 4. Ingestion rate (mg h^{-1}) of *Spisula subtruncata* as a response to different concentrations of total particulate matter (TPM: mg l^{-1}) and at different levels of organic matter. Mean + standard deviation.

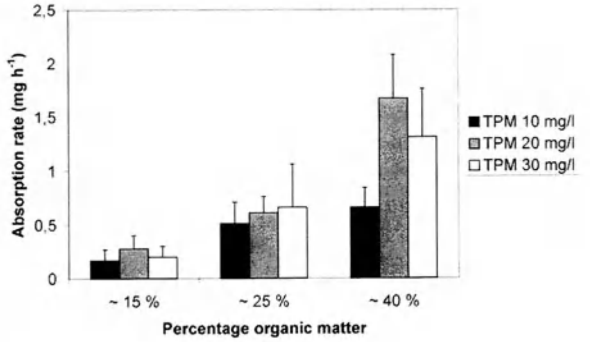

Figure 5. Absorption rate (mg h^{-1}) of *Spisula subtruncata* at different concentrations of total particulate matter (TPM: mg l^{-1}) and percentages of organic matter. Mean values + standard deviation.

not with the TPM (Table 2D). The equation relating AR with quality of the seston is AR = $-0.53 + 0.04\%\text{OM}$ (R^2=0.65; N=111; p<0.01) (Fig. 5). Absorption efficiency values ranged from 0.3 to 0.8.

Discussion

Many studies have been carried out relating food availability and filtration rate in marine bivalves, using seston concentrations below the threshold of pseudofaeces production (Navarro & Winter, 1982; Bayne et al., 1989) or above this threshold (Iglesias et al., 1992; Navarro et al., 1992; Bayne et al., 1993; Hawkins et al., 1996). The present study showed that filtration rate (FR) increased with seston concentration and decreased with higher values of food quality. Some authors (Iglesias et al., 1992; Navarro et al., 1992, 1994; Navarro & Widdows, 1997) also found an increase of FR with values of concentration of the seston (from 1 to 300 mg l^{-1}) in another infaunal bivalve such as the cockle *Cerastoderma edule* (Linné, 1758). Similar behaviour was observed in *Mytilus edulis* (Linné, 1758) by Bayne et al. (1993). In *S. subtruncata* the filtration rate increases with TPM but the increment is lower at higher food quality. This same effect has been described in *C. edule* both at low (Iglesias et al., 1992) and high (Navarro & Widdows, 1997) seston concentrations. A high FR is an efficient mechanism to enhance the energy gain from a turbid environment, because the species can process large amounts of particulate matter. *S. subtruncata* have the capacity to sort and preferentially ingest organic particles, and reject a high proportion of inorganic matter in the form of pseudofaeces.

Rejection of pseudofaeces has been described as a regulatory mechanism of ingestion rate (Iglesias et al., 1992; Navarro et al., 1992). Hawkins et al. (1996) found that the production of pseudofaeces in *M. edulis* depends on the FR, which increased with seston concentrations up to 112 mg l^{-1}. Navarro & Widdows (1997) obtained similar results for *C. edule* indicating an increase of the rejection rate with seston concentrations and a similar pattern for FR. In our study we have observed the same trend for *S. subtruncata* with an increase of RR at higher seston concentrations. At high quality of the diet there was a decrease of the rejection rate of *S. subtruncata*. Navarro & Widdows (1997) indicated that *C. edule* shows a mechanisms for regulating ingestion by increasing the rejection rates at high levels of FR but this mechanism can be modified by the quantity and quality of the suspended particulate matter. Iglesias et al. (1992) and Navarro et al. (1992) concluded that the mechanism of regulation in *C. edule* is determined by food composition, with reductions of clearance rate as the most important regulatory mechanism under high quality diets, and the production of pseudofaeces playing that role under low quality conditions. In *S. subtruncata* as an effect of filtration and rejection rate, ingestion rate (IR) was stable at different levels of organic matter content or seston concentration. Apparently increasing RR at different food conditions regulates IR. This regulatory mechanism has been described by Bayne et al. (1989) in the mussel *M. edulis*. They showed that maximisation of the ingestion rate occurred above the threshold of pseudofaeces production.

It is well established that bivalves are able to sort particles using their labial palps (Kiørboe & Møhlenberg, 1981; Newell & Jordan, 1983; Newell *et al.*, 1989, Ward et al., 1991) resulting in the preferential rejection of inorganic material in pseudofaeces. In *S.*

subtruncata the selection efficiency (SE) increased at high organic matter contents of the diets, although some negative values of SE occurred in experiments with diets containing low organic content due to mucus rejection. A pre-ingestive mechanism represents an adaptation to conditions with different quality of the diet. The trend recorded in our study is similar to other previous experiments with *C. edule* (Iglesias et al., 1992; Urrutia et al., 1996) but in our case lower values were obtained for similar organic contents. Iglesias et al. (1992) assumed that the rejected material by *C. edule* at food qualities below 40% of organic matter contains approximately constant fractions of mucus and that pseudofaecal mucus loss could be the explanation of the lower values of SE. These same authors suggested that the loss of secreted mucus in the pseudofaeces would be similar to the metabolic faecal losses associated with digestive processes.

Absorption rate (AR) in *S. subtruncata* was positively correlated to organic content of seston and no dependence was found with seston concentration. This is a result of both preingestive and digestive mechanisms at different seston qualities. Preingestive selection gives high OIR. This may be absorbed by *S. subtruncata* with higher efficiencies at high organic content as recorded in other bivalves (Navarro et al., 1994; Iglesias et al., 1996; Navarro & Widdows, 1997; Wong & Cheung, 1999). This increment of the efficiency of absorption at high food qualities has been described in *C. edule* and related to: (1) the rate of loss of endogenous organic components through the metabolic faecal losses (Ibarrola et al., 2000), (2) to the fact that the organic enrichment of the diet could result in a higher incorporation of food particles in the digestive gland indicating that more space becomes available for ingested particles (Navarro & Iglesias, 1993) or (3) that high quality particles experience a greater volume reduction when submitted to digestion as more space for incoming particles becomes available (Willows, 1992). Navarro et al. (1994) have indicated a variation of the gut passage time of the food in cockles with similar values of ingestion rates and at different contents of quality of the seston as it occurs in our study. This results in an increase of the absorption efficiency at higher quality of the seston with a significant relation of gut passage time with absorption efficiency.

Our results suggest that at different seston concentrations, *Spisula subtruncata* compensates efficiently by maintaining an effective preingestive mechanism of selection for organic particle matter, as well as increasing filtration and rejection rates for maintaining a constant ingestion rate. As a consequence, this species is well adapted to survive in environments with fluctuations in seston quality and quantity and maximizing the energy gain at high levels of organic matter in the water column caused by algal blooms in the coastal waters and a lower resuspension of fine sediments compared with estuarine waters.

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E. Orive, M. Elliott & V.N. de Jonge (eds), Nutrients and Eutrophication in Estuaries and Coastal Waters.

The management of nutrients and potential eutrophication in estuaries and other restricted water bodies

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Abstract

Conceptual models are derived to indicate the signs and symptoms inherent in nutrient changes to brackish, estuarine and coastal areas of restricted circulation. These give a structured approach to detecting adverse symptoms of hypernutrification and eutrophication at all levels of biological organisation, from effects at cellular levels to the ecosystem approach. The conceptual models illustrate the bottom-up approaches to the detection and control of potential problems and the importance of top-down responses. The bottom-up approaches incorporate mechanisms with regard to inputs, retention of nutrients, biogeochemical cycling and the primary production response. The top-down approaches include the detection of responses in high-profile components of the marine system, such as fisheries, sea mammals and wading birds and seabirds, which are often of paramount socio-economic or conservation importance. The management of the above causes and consequences, and following from the adoption by signatories to proposals given by the Paris Commission (PARCOM), can be accomplished by the derivation of Ecological Quality Objectives (EcoQO) and Ecological Quality Standards (EcoQS). These are given here as a development from the Environmental Quality Objectives and Standards (EQO/EQS) approach. Such EcoQO and EcoQS are regarded as as an aid to monitoring and management of estuaries and coastal waters. That management includes recent proposals within European legislation aimed at monitoring and managing the health and integrity of coasts and estuaries, for example the implementation of the Nitrates, Species & Habitats, and Water Framework Directives. The paper, therefore, discusses both the quality and quantity of data involved in the science required by managers and the way ahead for assessing and managing the fate and effects of nutrients. Using European and U.S. examples, the paper introduces the major challenge of how the concerns highlighted can be addressed by policy action.

Introduction

National and international bodies are increasingly required to consider the causes and concerns relating to eutrophication in the estuarine, coastal and marine environment and to provide management responses. This requires a structured approach by, firstly, defining the problem to give the basic understanding, the concerns by the public and the scientific and marine environmental management community and by determining the source of the problem. Secondly, the diagnosis of ecosystem pathology can both describe and assess the presence and severity of effects. Management tools for addressing these include the adoption of objectives and standards, the development of reference conditions and indicators to determine if those conditions are met, monitoring protocols and, eventually, the ability to have an increasing predictive capability using conceptual and numerical models. Thirdly, countries both singly and in tandem have to develop and implement policy strategies for addressing the problems although, as shown below, these are merging and leading to overlap. Those strategies should include controls on nutrient inputs.

The effects of excess nutrients in freshwaters have been well understood and documented since the mid-

20th Century. In contrast, an awareness of the potential importance of eutrophication in the coastal and marine environment has been developing only since the early 1980s (de Jonge & Elliott, 2001). Since then, the scientific determination and understanding of eutrophication and how it might be controlled through policy and regulation started to be incorporated in coherent frameworks both in Europe, through the Paris (OSPAR), Helsinki (HELCOM) and Barcelona Conventions, and in the U.S.A. through the National Oceanic and Atmospheric Administration (NOAA). Overall budgets of nutrients have been calculated for certain sea areas (as shown in the Quality Status Reports produced by OSPAR) (OSPAR Commission, 2000) although these reflect the adequacy of the data, which are good for point sources but poor for atmospheric and other diffuse sources.

A review of the available information (e.g. de Jonge & Elliott, 2001) indicates that whereas many scientific studies research in detail the causes and initial effects of the eutrophication, public and nature conservation concerns relate to high profile events. The latter include fish kills, closed fisheries, the reduced biodiversity of habitats, seabed de-oxygenation, reduced aesthetic quality of the coastal environment and even complete ecosystem collapse over extensive fjords and seas. These can be regarded as a set of causes leading to primary and secondary effects (Table 1). In turn, because of these events and public perceptions, scientists, policy makers and regulators are required to take an holistic view towards understanding the problems and producing remedial actions (see below).

Defining the problem

In addressing any problem, it is necessary to define and agree the terms used. The OSPAR Strategy to Combat Eutrophication (OSPAR, 1999) defines eutrophication as "*the enrichment of water by nutrients causing an accelerated growth of algae and higher forms of plant life to produce an undesirable disturbance to the balance of organisms present in the water and to the quality of the water concerned, and therefore refers to the undesirable effects resulting from anthropogenic enrichment by nutrients as described in the Common Procedure*". This definition, therefore, not only gives the cause but also the symptoms of effect. If it is accepted that aquatic systems have an assimilative capacity, i.e. the ability to absorb change or inputs before impacts are seen, then the definition of eutrophication by Schramm & Nienhuis (1996) is also of relevance: "*the process of natural or man-made enrichment with nutrient elements, mainly of nitrogen and phosphorous, beyond the maximum critical level of the self-regulatory capacity of a given system for a balanced flow and cycling of nutrients*".

The above definitions differ from that of hypertrophication (nutrient pollution) which may be regarded as "*over-enrichment or excess supply of nutrients beyond the maximum critical self-regulatory level to an extent that detrimental processes cause irreversible changes in aquatic communities, as long as nutrient levels are not reduced*" (Schramm & Nienhuis, 1996). In turn, this differs from the term hypernutrification which is regarded here simply as "*nutrient contamination – an excess of nutrients being present without adverse effects being manifest, the latter being the result of some other limiting factors*". The latter condition is particularly important and evident in estuaries which will have high levels of nutrients but those cannot be used to provide excess algal growth due to the high turbidity producing light limiting conditions.

Conceptual model of causes and effects

The actual and potential problems of nutrients and eutrophication can be defined and considered with reference to 'symptoms of ecosystem pathology', as the primary and secondary symptoms of change. These are used to produce a conceptual model of cause and effect and to indicate generic as well as site specific responses both according to geographical scale (extent of effects) and duration. The conceptual model can be considered as linking bottom-up causes versus top-down consequences (Fig. 1a, b). The former includes the physical nature of the system, the input levels and the initial biogeochemical cycling, whereas the top-down consequences include the effects on the macrobiological system: the macroinvertebrates, macrophytes, fishes and birds. As such, the higher order (top-down) effects may be more easily observed both by scientists but also by the public. Such changes, for example fish kills due to anoxia, therefore have a higher public perception but the understanding of those responses relies on knowledge of the bottom-up causes. As indicated above, the main problems occur in areas of restricted water exchange, such as the Baltic and Black Seas, fjordic and lagoonal systems, and those areas into which nutrients have been

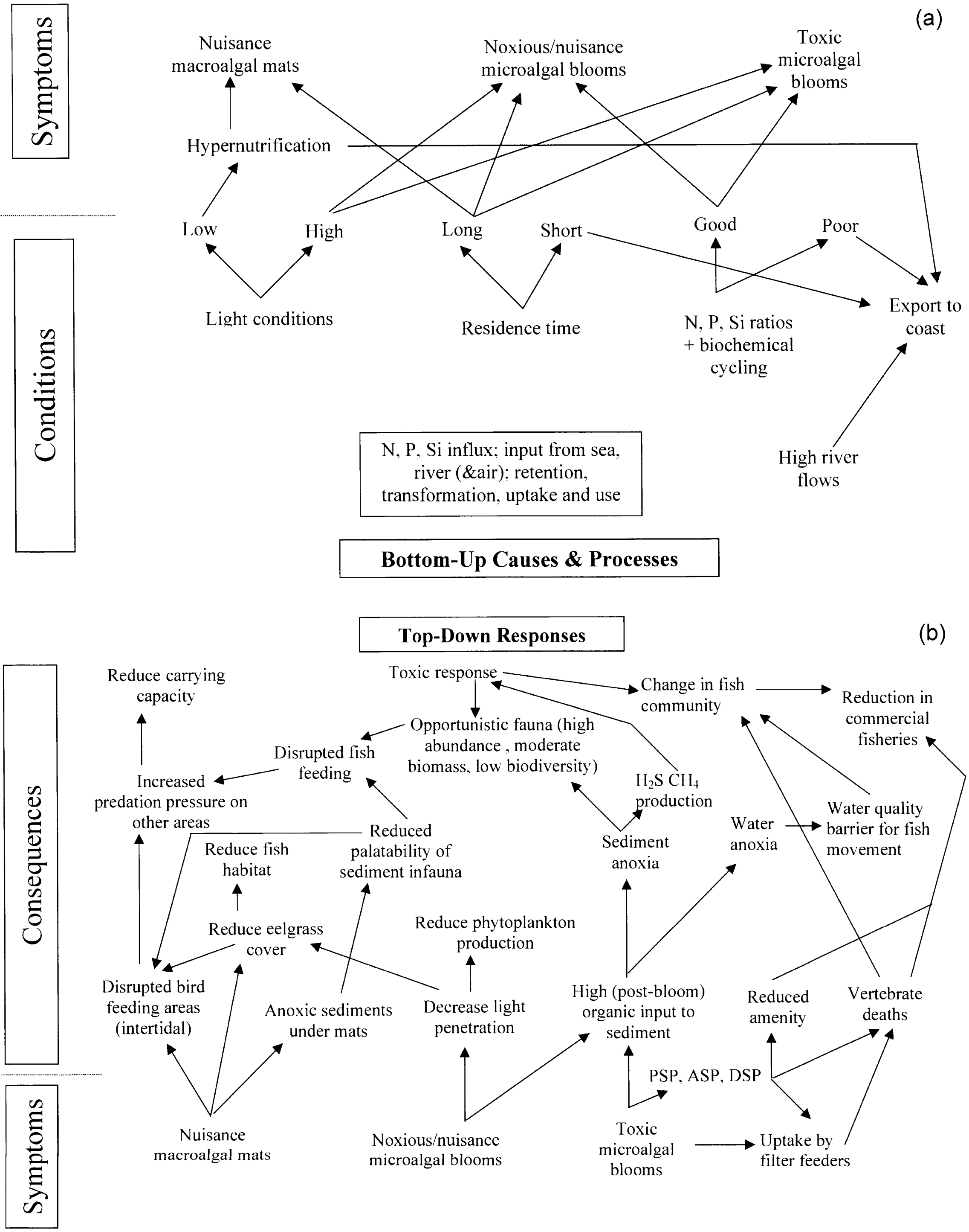

Figure 1. (a) Conceptual model – bottom-up causes and processes for nutrient effects. (b) Conceptual model – top-down responses for nutrient effects and eutrophication (adapted from Elliott & Hemingway, 2002).

Table 1. The causes, primary and secondary effects of eutrophication (developed extensively after an original format by Schramm & Nienhuis, 1996)

Causes	increased nutrient inputs; high residence time/slow flushing rate.
Primary effects	occurrence of blooms of toxic or tainting phytoplankton forms; increasing plant/algal biomass production; occurrence of blooms of micro-algae which may be a nuisance (and cause aesthetic pollution) through foaming (e.g. *Phaeocystis*, *Chaetoceros socialis*); decline or disappearance of certain perennial plants, often replaced by annual, fast growing opportunistic species such as foliose or filamentous green algae (e.g. *Ulva*, *Enteromorpha*); reduced diversity of the flora (and associated fauna); changes to photic regime through shading.
Secondary effects	increased particulate and dissolved organic matter in seawater and sediments; nuisance mat formation to hinder fishing and navigation; nuisance mat formation producing anoxic conditions; increase in microbial community and thus oxygen depletion, leading to hypoxic processes such as H_2S and CH_4 production; development of opportunistic macrobenthic populations and thus changes along the Pearson-Rosenberg continuum; poor water quality, especially water column oxygen depletion, thus affecting fishes and zooplankton; mortalities of higher organisms through effects of neuro-toxins; hindrance to intertidal feeding by wading birds and ducks.

transported from many sources. The latter includes the Skaggerak which receives nutrients in water transported from the North Sea states and atmospherically from the British Isles.

Attributes for the diagnosis of ecosystem pathology

In general, and for wide-scale application, changes in marine ecosystems as the result of human activities can be summarised as seven indicators (developed from Harding, 1992), many of which are reflected in concerns about nutrient enrichment (added in parentheses):

1. primary production (excess micro- and macro-algae),
2. nutrients (and their fate & effects),
3. species diversity (e.g. producing abiotic areas),
4. community instability (changing biotic composition, presence of opportunistic algae and invertebrates),
5. size and biomass spectrum (loss of ecosystem large elements, e.g. fish),
6. disease/anomaly prevalence (e.g. hypoxia-induced ailments in fishes),
7. and contaminant uptake and response (e.g. uptake of toxins).

In relation to the effects of nutrient enrichment, the characteristics of concern include, amongst others: the turnover time, including residence time and flushing rate leading to nutrient retention processes; the broad-scale physiography, its closed or open nature and its seasonality; the dominant physical processes (wind/wave/tidal/density currents, freshwater inflow); the high/low energy nature of the areas producing the hydrographic-sedimentary interrelationships; the biogeochemical processes in relation to hydrographical processes (N, P, C, Si cycling/dynamics); the depositing potential leading to the development of opportunistic populations via organic enrichment; the underlying control on the biological processes, e.g. the effect of turbidity on nutrient depletion; the nature of the area for supporting algal mats and the likely development of toxic or noxious blooms and their consequences; the value for higher predators and their response to eutrophication symptoms; and the carrying capacity for fishes (nursery, migration and feeding grounds) and birds (e.g. overwintering area).

Consequently, in order to determine and understand the extent and duration of the problem, there is the need for a reference framework and methods for defining reference conditions (as also indicated in the new EU Water Framework Directive). Reference conditions can be determined by one of 4 ways: hindcasting (using historical records), a direct comparison with another (pristine) area, predictive modelling, and the use of 'best-guess'/expert opinion. Hence, there is the need for historical budgets against which to compare the present situation as well as and widescale surveillance. However, it is emphasised that the latter should be monitoring focussed on the detection of the above signs and symptoms of nutrient enrichment effects.

Spatial occurrence of eutrophication

The location of eutrophic or potentially eutrophic areas requires to be determined in order to put remedial strategies in place. For example, within the U.K., OSPAR identified very few areas as of concern for eutrophication: notably the Ythan Estuary, Northeast Scotland, and Langstone Harbour, Hampshire, both of which were identified because of excessive intertidal macroalgal mats. Similarly, Seal Sands in the Tees estuary, north-east England, has increasing algal mats such that, as with other intertidal areas, there is concern regarding habitat integrity and the effect on bird and fish feeding as well as under-mat anoxia. Each of these cases reflects the retention of nutrients within the estuaries and harbours. However, in general, and in contrast to large areas of the Baltic and Skaggerak which develop extensive floating algal mats, most U.K. areas are sufficiently well-flushed such that nutrient enrichment does not present a problem. Despite this, other symptoms shown in the U.K. include hypernutrification and dissolved oxygen sags in many estuaries, especially at their turbidity maximum zones, the presence of *Phaeocystis* blooms off the Welsh and Irish coasts, and toxic microalgal blooms off the Scottish coast. It has been speculated that the latter were caused by nutrients from fish farming (MacGarvin. 2000).

The eutrophication signs and symptoms shown in the British Isles are considered here to be minor compared to other areas. For example, the Skagerrak and the Baltic areas have shown a transition from eelgrass (*Zostera*) domination to algal (*Cladophora* and *Enteromorpha* spp.) domination as the result of nutrient enrichment. This in turn reduces dissolved oxygen levels, plaice nursery areas and benthic biodiversity. Similarly, the Black Sea suffered an ecosystem collapse through eutrophication as did the Wadden Sea and Skaggerak/Kattegat in the late 1980s with a bloom of the microalga *Chrysochromulina* (see WHOI, 2001 for maps showing areas affected).

Environmental and socio-economic concerns

The major stakeholders in the marine and estuarine environment have concerns based on their desire for a healthy environment, without an increase in organic enrichment or its consequences. Conversely, and potentially as importantly, there are now also concerns by nature conservation bodies regarding decreases in organic enrichment, especially given the large landclaim of wetlands which previously supported large macrophyte populations and thus would have added detritus to support the estuarine detritus-based foodchains (Elliott & Hemingway, 2002). There are also concerns by nature conservationists regarding the potential loss of habitat (water, seabed and intertidal areas) and its integrity through increased enrichment. If such changes persist then ultimately there is a change in carrying capacity, especially the number of higher predators such as birds and fishes supported by an area.

In addition to the environmental consequences, and under the philosophies of Integrated Coastal Zone Management, it is necessary to take note of changes affecting other uses and users. Increasingly, environmental protection bodies, such as the Environment Agency in England and Wales, are required to incorporate socio-economic aspects into decision and policy-making. Similarly, it is axiomatic that the public and user-groups most often become most concerned when there are socio-economic repercussions of environmental change. The economic repercussions of eutrophication arise from their potential effects and impairment of use (Table 2); the latter may be due to actual or perceived economic importance and consequences (Bricker et al., 1999).

It is necessary to determine near and far-field effects, thus although one habitat such as an estuary may be affected, its role in acting as a sink by re-

Table 2. Socio-economic repercussions of eutrophication symptoms (modified from Bricker et al., 1999)

	Loss of habitat	Increased algal toxins	Fish kills	Offensive odours
Commercial fisheries	✓	✓	✓	
Recreational fisheries	✓	✓	✓	
Tourism	✓	✓	✓	✓
Human health		✓		
Water sports		✓		
Aesthetic values			✓	✓

ceiving nutrients from upstream as well as acting as a source to the coastal areas requires to be considered. Furthermore, there is the need to quantify the aspects which are quantifiable in economic terms, such as fisheries, but also to quantify the 'un-quantifiable' aspects such as aesthetical characteristics and values. For example, whether it is possible to value the degradation of an area when perceived by those using it for recreation. Similarly, there is the need to determine the value of possible increased fisheries, with the input of organic matter, but also the costs of decreased fisheries/habitat where environmental degradation occurs. There may also be costs due to the losses in any nutrient (cost) budget, i.e. the wasteful or ineffective use or rapid removal from the land of fertilisers. Finally, it is necessary to determine the costs of treatment, mitigation and/or compensation in addressing the problem. As yet, there has been no comprehensive assessment of these economic aspects although the ongoing EU Framework V research programme EUROTROPH will carry this out for a few estuarine and coastal areas (see EUROTROPH, 2001).

Objectives, standards, reference conditions and monitoring

In common with tackling other marine problems, it is necessary to define the habitats at risk and to decide whether to assess all habitats or to prioritise and thus only monitor where problems are likely. This is particularly difficult in the case of diffuse causes of pollution where the causes and thus the effects may be widely dispersed. Then it is necessary to carry out targetted monitoring against a set of indicators which assess specific symptoms. This is regarded here as monitoring *sensu strictu* and thus is distinguished from surveillance; in the latter, field surveys are carried out with a *post hoc* detection of trends followed by the explanation of those trends. The resultant indicators may be derived and used singly or in combination and on a spatial (extent) and/or temporal (duration) basis, on a taxonomic (taxon-specific) and/or non-taxonomic (e.g. environmental) basis, or on a structural (i.e. characters at a single census point) and/or functional (rate processes) basis. This approach includes the setting of objectives and standards (Elliott, 1996) and testing for compliance with them. Hence, the move by the EU Directives and OSPAR towards considering Ecological Quality (EcoQ) and Ecological Quality Objectives (EcoQO). It is suggested here that EcoQO are accompanied by Ecological Quality Standards (EcoQS) as a further tool to be used as a basis for monitoring (see below). However, while such tools, including indicators, attempt to look for a change as a response to nutrient inputs, it is necessary, especially in variable environments such as estuaries, to consider that an environment can have a large capacity for absorbing nutrients before effects are manifested. Such a resilience can be regarded as *environmental homeostasis*.

As an approach to the monitoring and management of waters, and as a precursor to the EcoQO/EcoQS debate, the U.K. has long adopted the EQO/EQS (Environmental Quality Objectives/Standards) approach whereby *objectives* are statements and *standards* are numerical values. EQO/EQS are also incorporated into certain EU environmental protection Directives (Elliott et al., 1999). These have been derived and then used for determining use-related achievement and for chemical and microbiological determinands, e.g. as shown with compliance for the EU Bathing Beaches Directive. Any objective created, as the desired aim for an area such as the ability to maintain an activity such as bathing, requires to be accompanied by one or more standards, for example the levels of coliform bacteria. They should provide reference conditions or an end-point against which effective monitoring is performed such that it is difficult to create monitoring programmes or to see if an environmental quality objective is met unless standards are produced. Consequently, the approach requires monitoring protocols to be rigorously defined.

Under the ecosystem approach now adopted widely, these objectives and standards are now required to address nutrient and eutrophication problems. For example, an objective may be to prevent macroalgal mats forming on intertidal areas because of the resulting effect on the top predators such as fish and wading birds. The latter have a high public and nature conservation relevance. The accompanying standard or reference could be that no more than x% of mudflat should be covered by opportunistic green algae during later summer periods. Similarly, if an adopted quality objective is to allow fish to migrate through an estuary then the accompanying standard could be that dissolved oxygen has to exceed 5 mg l^{-1} (Elliott & Hemingway, 2002). In each case, the monitoring is then designed with sufficient rigour and frequency to detect such a change and management actions are then taken if the standard is breached.

EcoQO, rather than EQO, becomes a more realistic term for addressing the problems caused by nutrient enrichment given that biological and ecological health-related objectives are required. There is the need to separate these from true EQO, i.e. those relating to the human use of the system such as the requirement for an area to support bathing. The extension of EQS to EcoQS requires a large degree of development, for example, whereas EQS are usually derived for chemicals (DO, NH_4^+, trace metals), EcoQS are required for ecological or biological health variables. Despite this, there are few examples of EQO and EQS in legislation and statutory frameworks: the U.K. Water Act 1989 provides for statutory objectives and standards but only those related to European Directives have legal status; the U.K. environment protection agencies have locally adopted such standards, and the EA in England and Wales uses non-statutory estuary quality objectives as a basis for water quality management and licensing. Similarly, the OSPAR signatories agreed at Sintra, 1998, to achieve concentrations near background levels for natural substances and at zero for synthetic substances – this is also a use of EQS, whereby discharge controls and monitoring are directed towards achieving the standards. Finally, it is of note that the Dutch RIKZ has adopted operational objectives towards developing criteria for judging eutrophication, its causes and consequences.

Models as tools for nutrient management

Conceptual, analytical and numerical models help to identify, explain and tackle problems; for example the conceptual models here (Fig. 1a, b) summarise the scientific and conceptual framework and thus they are valuable in presenting the potential problem to managers and policy-makers (Read et al., 2001). In turn, quantifying the links in conceptual models will produce numerical models. There is a hierarchy of the latter ranging from hydrodynamic models (of water movements, flushing rates, etc, and which may be two and three dimensional) to biogeochemical models (e.g. giving chemical processes such as nutrient transformations), population models (showing individual number change such as in microbial or fisheries populations), community models (allowing the prediction of community structure) and finally to ecosystem models such as GEMBASE and BOEDE which aim to summarise ecosystem structure and functioning. However, with progression through this ecosystem hierarchy there is increasing inherent variability (i.e. 'noise') such that it is more difficult to observe a 'signal' as a measure of change, a greater uncertainty of prediction, and a poorer understanding of processes (Read et al., 2001). All of these dictate that there is likely to be less willingness for their use in management.

Numerical models can be stochastic and deterministic in that they reflect the underlying science and involve equations determined by the physics and chemistry interrelationships, for example the exchange between ammonia, nitrate and nitrite depending on the prevailing conditions. In contrast, the models can also be empirical in that they reflect observed patterns and trends but without being based on any theoretical basis. The latter may be based on regression or multiple regression equations which link independent environmental variables, such as water transparency and nutrient concentrations, with a biological (response) dependent variable such as chlorophyll *a* content (Read et al., 2001). With increasing data and a better understanding, the adequacy and predictive capability of these models may increase but again their value in reaching management decisions is still questionable.

Policy strategies – development, merging & overlap

Since the middle of the 20th century, there has been a sequence of policy action in defining and tackling the marine environmental problems of developed countries. In the 1960s, it was realised that there is a problem of pollution; in the 1970s that there was no problem as dilution was a solution (via long-sea outfalls); this was then regarded as being insufficient in the 1980s, and hence end of pipe controls were increased.

In the 1990's it was accepted that the problem was not just related to pollution *per se*, was much larger than acknowledged and that an holistic and ecosystem approach was required together with industrial Environmental Management Systems. At the start of the 21st century, it has been realised that most of the previous solutions relate to the more-easily tackled point-source pollution whereas the problems now mainly relate to diffuse pollution. As such, catchment and open sea solutions and strategies are required. This progression of different strategies was accompanied by a movement from a sectoral approach, in which each activity (pollution, fisheries, coastal defence, etc.) is addressed separately, to a multi-sectoral approach. In the latter, there is the need and desire to consider all activities concurrently, hence the ecosystemic holistic approach.

In essence, it has been accepted that point-source pollution is addressed relatively easily, albeit expensively, whereas diffuse pollution requires a greater degree of co-ordination and wider-scale thinking. Accompanying this sequence of addressing concerns, there have been many initiatives and developments (Table 3) which have some common themes but also have led to confusion and overlap in marine environmental management in NW Europe (Ducrotoy & Elliott, 1997). An increasing number of policy initiatives set the framework and priorities to be addressed, they require the creation of objectives and standards, and they are designating areas for protection and assessing the condition of that area. Some of these initiatives are source-orientated, in that they address the cause of change, whereas others are target-orientated in that they address the effects in biological components.

Because of the actual or potential duplication (see also Ducrotoy & Elliott, 1997), there is a recommendation here of the need to bring together the Oslo and Paris Commission, the EU Directives (for Water Framework, Integrated Pollution Prevention & Control, Habitat & Species Protection, Urban Wastewater Treatment (UWWT), and Nitrates), the International Council for the Exploration of the Sea, and the North Sea Ministerial Conference. In bringing these together, there is the opportunity to remove anomalies in the systems. For example, there is a spatial conflict in areas under control and in procedures which is shown by the limits covered by OSPAR and the Habitats & Species Directive (to the 200 nm limit) and the 1 nm covered by the Water Framework Directive (unless this eventually becomes extended). This is despite the fact that the latter Directive purports to give added protection to areas already protected under other Directives.

Tackling the problem (1): OSPAR in NW Europe

The OSPARCOM has agreed a set of strategies: for the *Protection and Conservation of Ecosystems and Biodiversity (P&CE&BD)*; to *Combat Eutrophication*; to prevent the discharge of *Radioactive Substances* and *Hazardous Substances*; and for the control of *Offshore Oil and Gas* developments and impacts. Within these, it has identified a set of 19 issues of which four have been adopted as the most important: benthic communities, planktonic communities, nutrient budget and production, and oxygen consumption, each of which can be linked to the primary and secondary symptoms of eutrophication (Table 4). It has agreed to use a target-orientated approach (i.e. as an ecological response) as well as a source-orientated approach (reduction in diffuse and point sources). In the terms used above, these are, respectively, top-down responses and bottom-up processes and causes.

As nutrient transport and thus eutrophication are trans-boundary problems, OSPAR has included them as one of their strategies with a general objective – '*to achieve and maintain a healthy marine environment where eutrophication does not occur*'. The strategy is to define problem areas, potential problem areas or non-problem areas, to develop EcoQO by 2002 and to produce a healthy marine environment by 2010. This in turn requires management tools such as indicators and targets as well as adequate and effective monitoring. Hence, OSPAR has adopted a Common Procedure to identify the eutrophication status of marine areas which requires the implementation of integrated target-orientated and source-orientated actions for problem areas. These include determining EcoQO which OSPAR regard as '*the desired level of biological quality relative to the reference level*'. Hence, as interpreted here, there is a requirement by member states, following the Common Procedure, to develop and adopt EcoQO and their accompanying EcoQS for the components at risk (as early warnings of change or the end point of change). Under this rationale, EcoQS can be interpreted as reference levels and as such they overlap with Reference Conditions identified in the EU Water Framework Directive and the Favourable Conservation Status identified in the EU Habitats & Species Directive (Elliott et al., 1999).

Table 3. The OSPAR proposed set of issues translated into the appropriate eutrophication symptoms and signals and the ecosystem at risk

Proposed set of issues	Relevance to eutrophication symptoms/signals	Ecosystem at risk
Reference points for commercial fish species	interference with migration routes	estuaries
	interference with nursery areas	estuaries/sandy beaches
	interference with feeding and refugia	mudflats, saltmarsh
Threatened or declining species	prevention of migration (e.g. shads)	upper estuarine areas
Sea mammals	production of toxic blooms	open coastal
Sea birds	production of toxic blooms	open coastal
	interference with feeding by algal mats	intertidal mudflats
	change in palatability of prey through anoxia	low energy areas
Fish communities	change to herbivores	inshore areas
	interference with use	all marine areas
Benthic communities	movement along Pearson-Rosenberg (Rhoads-Germano) model	sedimentary areas in low energy regimes
Plankton communities	community change	inshore areas
	dominance of certain taxa	inshore areas estuaries
Habitats	habitat integrity	inshore and estuarine areas
Nutrient budget and production	hypernutrification	areas with poor water exchange
	increased input and retention	inshore, estuarine, fjords
Oxygen consumption	increased consumption	restricted circulation areas
	tendency to hypoxia then anoxia	stratified and restricted areas

Together with the need for this strategy to assess the extent of the signs and symptoms of eutrophication, a joint strategy is needed to control the causes of the change. In Europe, this is being tackled for point source discharges, such as via the Urban Wastewater Treatment Directive, and diffuse sources, such as via the Nitrates Directive and the creation of Nitrate Vulnerable Zones (Elliott et al., 1999). However, a difficulty faced by states such as the U.K., where the symptoms are relatively minor compared to elsewhere, is to convince politicians, policy makers, environmental managers and the public that diffuse pollution and the transport of pollution are problems to be tackled on a wide scale. For example, they have to acknowledge that the country contributes to problems experienced elsewhere, for example in Scandinavia.

Tackling the problem (#2): NOAA in the U.S.

The U.S. NOAA has recently produced a comprehensive means of addressing estuarine eutrophication problems (Bricker et al., 1999). It has defined primary and secondary symptoms and then given high, moderate and low scores for each symptom. This has been used to produce a matrix of the symptoms to give an overall expression of concern of eutrophic conditions. Finally, these in turn have produced a regional assess-

Table 4. NW European marine and coastal waters: controls on nutrient inputs and their effects (see text for abbreviations)

		Frameworks	Priorities	Objectives	Area designation	Condition assessment	Standards	Source orientated	Target orientated
1970	EQO/EQS			√			√	√	
1974	OS/PARCOM	√	√					√	√
1979	Wild Birds Directive	√			√	√			√
1991	Nitrates Directive	√		√	√			√	
1991	UWWT Directive	√			√	√		√	
1992	Habitats & Species Directive	√	√	√	√	√	√		√
1992	OSPAR	√	√					√	√
1995	NSeaMin Conference	√	√					√	√
1996	IPPC Directive		√				√	√	
1998	OSPAR Sintra	√	√					√	√
1998	OSPAR Eutrophication Strategy			√	√	√	√	√	√
1998	OSPAR P&CE&BD Strategies			√	√	√			√
2000	Water Framework Directive	√	√	√	√	√	√	√	√

ment, including an analysis of data completeness and reliability (DCR), and hence a national strategy. It is of note that the U.S. evaluation relied where possible on available data but that in many areas, especially those where little monitoring has been carried out, there are few data available. In those cases, the evaluation relied on local knowledge. As such, the approach used indicates that such an evaluation can be carried out using expert judgement where data are lacking and still produce a worthwhile result.

This detailed assessment concludes that symptoms of eutrophication are prevalent in U.S. estuaries; that there is substantial human influence on the expression of eutrophic conditions; that impairments to estuarine resources, and fisheries in particular, are of great concern; that management requirements depend on eutrophic conditions and susceptibility; that without preventative efforts, eutrophic conditions can be expected continually to worsen; that much effort is needed to improve the characterising and understanding of estuarine eutrophication; and that assessment results will be valuable in setting national priorities. While all of these are to be expected, they very well illustrate the value of a comprehensive and objective evaluation of the problem.

Concluding remarks – the way ahead: managing nutrient inputs, fate & effects

With respect to nutrients, any system may show the low level effects of change but within an inherent capacity for absorbing that change (regarded here as 'environmental homeostasis'). In management terms, it is better to take as the end point the aspects of particular socio-economic significance or those significant in human perception, e.g. the aesthetic aspects. Therefore, there is the need to separate causes and symptoms and to regard eutrophication as a set of undesirable symptoms. This includes separating it from hypernutrification (nutrient contamination) but ensuring that it is broader than organic enrichment. The definition and assessment of nutrient effects also have to allow for varying critical levels of inputs, for example in the spectrum between oligotrophic systems and naturally-eutrophic estuaries. It also has to encompass allowing for coastal areas receiving nutrients but where those nutrients are successfully used to enhance productivity without creating undesirable effects. In this, it is also necessary to acknowledge that most estuarine and marine systems are resilient and thus recovery is achievable with a reduction in inputs.

As shown here, it is necessary to consider our confidence in ascribing effects to eutrophication drivers.

There are some good case-histories of the consequences of nutrient inputs, the changes to primary producers and the changes at higher trophic levels. This qualitative approach has produced the conceptual models. However, there are insufficient case-studies, especially of holistic studies, to quantify the sequence of processes. Similarly, there are extensive modelling approaches but one questions whether these are yet sufficient for management at ecosystem level. Linked to this is the use of schemes for classifying marine waters (*a la* the EU Water Framework Directive) and indicators of change related to ecosystem pathology which can be used as management tools.

Recent initiatives, such as by OSPAR and the EU, require reference conditions to be determined based on the causes, signs and symptoms of eutrophication for major ecosystem types. An increasing area of confusion, however, is that these are variously termed trigger values, favourable status, references conditions, indicators or EcoQO/EcoQS/EQO/EQS. Irrespective of the term, they should be linked to monitoring protocols and strategies. Given the nature of the systems to be managed, site specific characters need to be retained but within a manageable set of objectives. This should include identifying those priority ecosystems at risk – by signs, symptoms and causes – which primarily will be inshore areas, areas of restricted circulation and areas likely to have a socio-economic importance or aesthetic, social significance.

As indicated here, there is the paradox that nutrients are required for ecosystem function but an excess of them produces a set of undesirable symptoms, termed eutrophication. Some areas have a high assimilative capacity for those nutrients, often by increased dilution or flushing, whereas others can hold nutrients without showing undesirable symptoms; a third set of areas both retains the nutrients and has physical conditions suitable for creating undesirable symptoms. The latter are the areas which attract attention from the managers and policy makers and thus where there is the greatest political will to address discharges. This has led to improvements by removing nutrients through discharge control.

A second paradox here is that such a perceived improvement in environmental health, i.e. the removal of anthropogenic inputs and excess nutrients, can lead to an overall deterioration in ecosystem functioning. In many estuaries, the removal of anthropogenic organic matter has been accompanied (or in some cases preceded) by a loss of wetlands, usually through land-claim. The latter has resulted in an often-unquantified reduction in detrital inputs, for example the reed-bed and saltmarsh areas lost through land-claim would formerly have been a major source of detritus. Hence, the loss both of natural detritus and anthropogenic organic matter in estuaries, which rely on the detrital pathways for their functioning, should cause concern. In a similar vein, catchment land use and agricultural practices in developed countries result in nutrients being retained less on the land, an increase in water removal from the land and thus greater flushing of estuarine systems. This may reduce the ability of estuaries to act as sinks for nutrients but increase their role as a source to adjoining coastlines.

Our conceptual understanding of such changes is good but less so our quantitative understanding. Despite this, we have the ability (but not necessarily the desire) to make policy decisions based on those inadequate data. The prime difficulty for policy makers and regulators is that whilst eutrophication effects can now be more effectively demonstrated in the coastal environment, the main (uncontrolled) input sources are predominantly diffuse – from land runoff and atmosphere. Hence, addressing these requires different approaches such as changing land practices and reducing combustion products.

It is concluded here that there are several areas for development in order to understand, address and communicate actual and potential problems due to nutrients:

Know the problem: there is the need for effort covering both widespread surveillance (low level) and identified and targeted monitoring;

Research needs: such as in the EU-LIFE ALGAE, MARE and EUROTROPH programmes which bring together the science, modelling and management aspects, including the socio-economic aspects of nutrient and organic reduction programmes;

Lateral thinking: the need for nutrient controls as well as other methods such as the recovery and sustainable use of algal mats (as in the EU-LIFE ALGAE programme where algae are used for paper, cellulose and biogas production as well as fertiliser);

Catchment management: there is the need for catchment nutrient budgets (*a la* the EU Water Framework Directive), and an acceptance of the repercussions of land-use changes such as fast land drainage and the poor land retention of nutrients;

Test the rationale: there is the need to further define the characteristics of the ecosystem, to define EcoQO and EcoQSs, to define the monitoring strategy and monitoring protocols and finally to determine compliance with each of these;

Question the need: for a detailed assessment involving a large data gathering exercise in contrast to the use of 'expert judgement'. i.e. to question what information is required by managers of designated areas or catchments;

Simplify the debate/approach: to get OSPAR to adopt the EcoQO/EcoQS distinction and to get the various bodies to harmonise terms and approaches across their initiatives.

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