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1 **Twenty years of research in ecosystem functions in aquatic microbial ecotoxicology**

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3 Soizic Morin and Joan Artigas

4

5 **Abstract**

6 One of the major threats to freshwater biodiversity is water pollution including excessive loads of
7 nutrient, pesticides, industrial chemicals, and/or emerging contaminants. The widespread use of
8 organic pesticides for agricultural and non-agricultural (industry, gardening, etc...) purposes has
9 resulted in the presence of their residues in various environments, including surface waters. However,
10 the contribution of pesticides to the deterioration of freshwater ecosystems (i. e. biodiversity decline
11 and ecosystem functions impairment) remains uncertain. Once in the aquatic environment,
12 pesticides and their metabolites can interact with microbial communities causing undesirable effects.
13 The existing legislation on ecological quality assessment of water bodies in Europe is based on water
14 chemical quality and biological indicator species (Water Framework Directive, Pesticides Directive)
15 while biological functions are not yet included in monitoring programs. In this literature review, we
16 analyse the last twenty years (2000-2020) of research on ecological functions provided by
17 microorganisms in aquatic ecosystems. We describe the set of ecosystem functions investigated in
18 these studies and the range of endpoints used to establish causal relationships between pesticide
19 exposure and microbial responses. We focus on studies addressing the effects of pesticides at
20 environmentally realistic concentrations and at the microbial community level to inform the
21 ecological relevance of the ecotoxicological assessment. This literature review highlights that most
22 studies were performed using benthic freshwater organisms, and that autotrophic and heterotrophic
23 communities are most often studied separately, usually testing the pesticides that target the main
24 microbial component (i. e. herbicides for autotrophs and fungicides for heterotrophs). Overall, most
25 studies demonstrate deleterious impacts on the functions studied, but this review points to the
26 following shortcomings: i) the non-systematic analysis of microbial functions supporting aquatic
27 ecosystems functioning, ii) the study of ecosystem functions (i. e. nutrient cycling) via proxies (i. e.
28 potential extracellular enzymatic activity measurements) which are sometimes disconnected from
29 the current ecosystem functions, and iii) the lack of consideration of chronic exposures to assess
30 impact, adaptations or recovery of aquatic microbial communities to pesticides.

31

32

33 **Keywords:** aquatic microbial ecotoxicology, community-level effects, microbial functions, pesticides.

34

35 **Running head:** Ecosystem functions in aquatic microbial ecotoxicology

36

37

38 **1. Introduction**

39 Aquatic microbial ecotoxicology (AME) is a research subject at the interface between aquatic
40 microbial ecology and ecotoxicology (Ghiglione et al. 2016). In aquatic ecosystems, microbial
41 communities cover a large diversity of microorganisms, living forms (planktonic/benthic,
42 solitary/colonial...), and functionalities. They are composed of autotrophic (including cyanobacteria,
43 green algae and diatoms, among others) and heterotrophic microorganisms (archaea, bacteria, fungi
44 and protozoa) and play a pivotal role in global biogeochemical cycles, nutrients cycles and energy
45 flow in aquatic ecosystems (Lock et al. 1984, Battin et al. 2003). One of the major goals of AME is to
46 improve mechanistic understanding of ecologically significant responses of microbial communities to
47 contaminants, and their potential impact on higher trophic levels in aquatic food webs. While various
48 studies have investigated the impacts of pesticides on the structure and diversity of aquatic microbial
49 communities (*e. g.* Debenest et al. 2010, Staley et al. 2015), information is still needed to understand
50 how pesticides affect ecosystem functions supported by microorganisms.

51 Leenhardt et al. (2022) proposed a framework using a set of clearly-defined core categories of
52 ecosystem functions and services supporting the identification of effectively or potentially
53 threatened function. For instance, microbial autotrophs are involved in ecosystem functions
54 production of organic matter (OM) and the provision and maintenance of biodiversity. They also
55 contribute to gas regulation given their photosynthetic character and to the cycling of nutrients.
56 Microbial heterotrophs play a pivotal role in the regulation of carbon and nutrient cycles, gas
57 exchanges with the atmosphere, propagule dispersion, and pollutant mitigation in the aquatic
58 ecosystem (Leenhardt et al. 2022). Both the autotrophic and heterotrophic components are involved
59 in the provision and maintenance of biotic interactions within the microbial community and through
60 the aquatic food web. The ecosystem function categories described in Leenhardt et al. (2022) are
61 based on the works by De Groot et al. (2002) and Pettoirelli et al. (2018) with some modifications.
62 Ecosystem functions supported by microbial communities can be directly measured or estimated
63 using a set of proxies which simplifies the methodological approach.

64 In this context, we performed a systematic review of pesticides' effects on the ecosystem functions
65 associated with aquatic microbial communities over the last twenty years. The main objectives of this
66 review were i) to characterise the main functions assessed and how they are impacted by pesticide
67 exposure and ii) to identify overlooked microbial functions and future research perspectives to better
68 consider the effects of pesticides on the ecosystem functions supported by microbial communities.

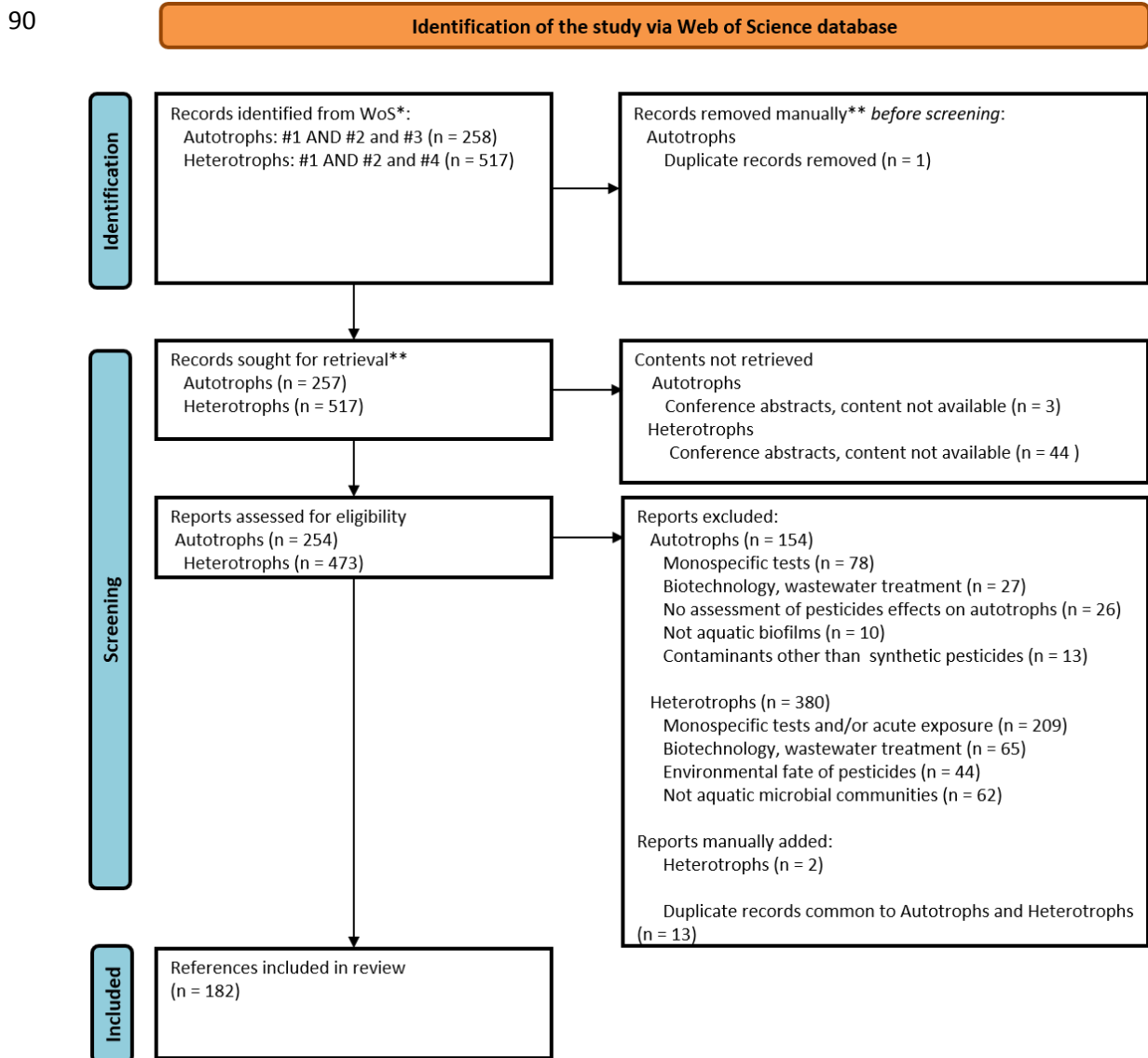
69

70 **2. Systematic review of AME studies addressing ecosystem functions: Methodology**

71 We retrieved all Web of Science publications (including original research articles, reviews and book
72 chapters) published in English between 2000 and 2020 to gather existing knowledge on pesticides'
73 effects towards the diversity and functions of aquatic microbial communities. The literature search
74 was performed on the Title and Author keywords, by combining requests (detailed in Supplementary
75 Information: S11) related to aquatic ecosystems (query #1) and pesticide exposure (query #2), with
76 the targeted microbial communities (query #3 for the autotrophic component or query #4 for the
77 heterotrophic component).

78 From these combinations of queries, we harvested a limited number of relevant references for the
79 autotrophic component of microbial communities. More specifically, 258 references were retrieved,

80 of which 61% were finally discarded (Figure 1). A total of 517 references were retrieved for the
 81 heterotrophic component. Among these 517 references, only 18% were retained (Figure 1). After
 82 clean-up, a complete list of 182 references were conserved for data analysis, where records related
 83 to autotrophic and heterotrophic components of microbial communities were balanced (Figure 1, see
 84 reference list in SI2) . We observed a very low overlap of references between queries made for
 85 autotrophic or heterotrophic components (7%) highlighting the fact that studies cover ecosystem
 86 functions supported by one or the other specific component of aquatic microbial communities.
 87 Overall, studies distinguish pesticide effects between autotrophic and heterotrophic components of
 88 microbial communities are few, although several functions can be ensured by both (i. e. respiration
 89 and nutrient uptake, among others...).



91 Figure 1. PRISMA 2020 flow diagram for the systematic review (Page et al. 2021). Detailed search
 92 requests are available in SI1 (*). No automation tools were used (**).

93 **3. Results and discussion**

94 **3.1. Communities targeted and experimental conditions**

95 The first outcome of our literature review was that most studies (72%) analysed the impacts of
 96 pesticides on communities living in biofilms rather than in planktonic habitats (Table 1). AME covers
 97 studies in all types of aquatic ecosystems; however, much more studies have been conducted in
 98 freshwaters (85% of the records) than in marine or transitional ecosystems (Table 1; see also Zhao et
 99 al. 2022). This trend observed in the literature review could be due to the dilution effect of pollutants
 100 in marine waters compared to freshwaters (Pesce et al. 2021; Leenhardt et al. 2022). Accordingly,
 101 most studies in the marine environment were conducted in the coastal zone close to sources of
 102 pollution. Among freshwater ecosystems, 56% of the studies dealt with lotic environments (streams
 103 and rivers) followed by studies in lentic environments (ponds, lakes and reservoirs, 28%). A
 104 consistently lower percentage of studies conducted in lentic environments was observed for
 105 heterotrophic components compared to autotrophic parts of microbial communities (Table 1).

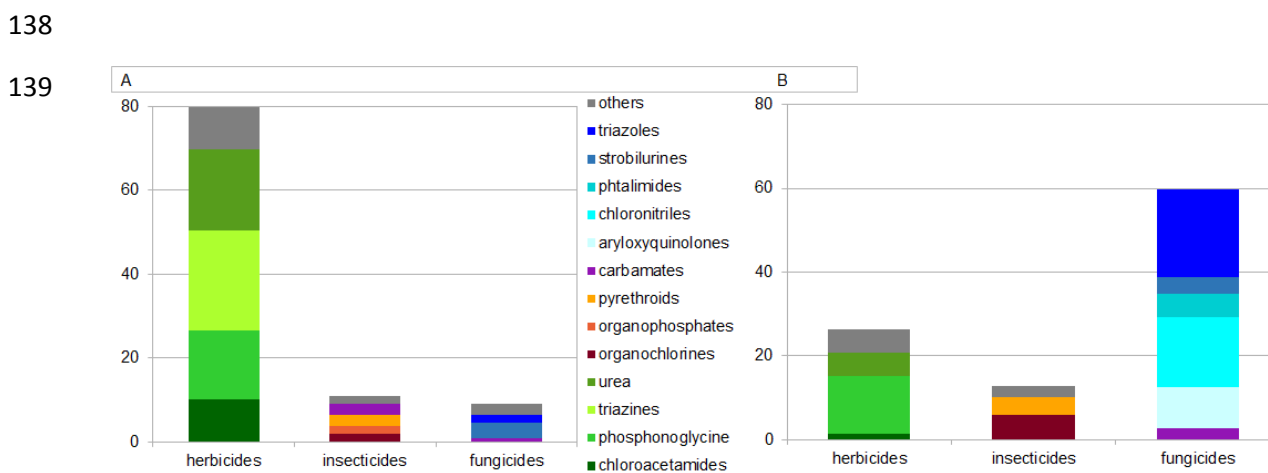
106 This literature review focusing on AME studies performed at the microbial community and ecosystem
 107 scales highlights that both experimental (nanocosms, microcosms and mesocosms, *sensu* Newman
 108 and Clements 2008) and field approaches (i. e. watershed) are used, but with some preferences:
 109 Table 1 indicates that 69% of AME studies were conducted at the community level in microcosms (i. e.
 110 test tubes, laboratory jars, small aquaria, etc...) followed by field studies and mesocosms (e. g.
 111 artificial flumes, ponds, enclosures) approaches. Comparatively, the use of microcosm approaches
 112 compared to field and mesocosm approaches is much more imbalanced when studying heterotrophic
 113 than autotrophic parts of the microbial communities. The strong reproducibility and increasing
 114 standardisation (e. g. OECD 2006) of micro- and mesocosm approaches make them a highly
 115 complementary tool for discerning the effects of pesticide in the natural environment. Despite
 116 efforts made by the AME researchers' community to better link laboratory to field results, recurring
 117 difficulties persist in filling the gaps (Vignati et al. 2007; Artigas et al. 2012a).

Number of records	Autotrophic part	Heterotrophic part
Habitat		
-Benthic (biofilm)	60	73
-Planktonic/pelagic	28	13
-Both habitats	7	5
Environment		
-Lotic	50	54
-Lentic	40	14
-Coastal/estuarine	5	23

Experimental approach		
-Microcosms/nanocosms	45	80
-Mesocosms	18	4
-Field	32	7

118 Table 1: Habitat, type of aquatic environment and experimental approach implemented in AME
 119 studies between 2000 and 2020 for autotrophic components (n = 95) and heterotrophic components
 120 of aquatic microbial communities (n = 91).

121
 122 Forty-five pesticides were considered in the references analysed, which is low compared to the large
 123 number of pesticides used and present in aquatic environments (e. g. Sharma et al. 2019). As
 124 expected, the mode of action of the pesticides tested was in line with the main microbial component
 125 studied (Figure 2). Articles targeting autotrophic components of aquatic microbial communities
 126 mostly addressed the impact of herbicides (60% of studies), in particular photosynthesis inhibitors
 127 (atrazine: 18 records, followed by diuron: 13 records, and isoproturon: 7 records) and the broad-
 128 spectrum herbicide glyphosate (phosphonoglycine, 18 occurrences). The highest diversity in
 129 herbicide molecules belongs to the chemical families of chloroacetamides (7 molecules dominated by
 130 (S-)metolachlor: 4 records over a total of 11), triazines (26 records for 5 compounds, dominated by
 131 atrazine) and ureas (diuron, isoproturon and chlortoluron totalizing 21 occurrences). In the case of
 132 heterotrophic components, the effects of fungicides have been addressed predominantly (61% of
 133 studies), followed by herbicides and to a lesser extent insecticides. However, a similar diversity of
 134 fungicide and herbicide molecules were tested. Tebuconazole (22 records, triazole) and
 135 chlorothalonil (20 records, chloronitrile) in the case of fungicides, and glyphosate (17 records,
 136 phosphonoglycine) in the case of herbicides, were the most studied molecules when dealing with
 137 pesticides effects on the heterotrophic component of aquatic microbial communities.



140 Figure 2: Chemical families of the 3 main classes of pesticides (bars = counts, 1 family may contain
 141 several molecules) tested against aquatic microbial communities under controlled conditions,
 142 excluding field and micro/mesocosm studies where complex environmental mixtures of pesticides
 143 were present. Note that several studies include 2 or more pesticide molecules, sometimes

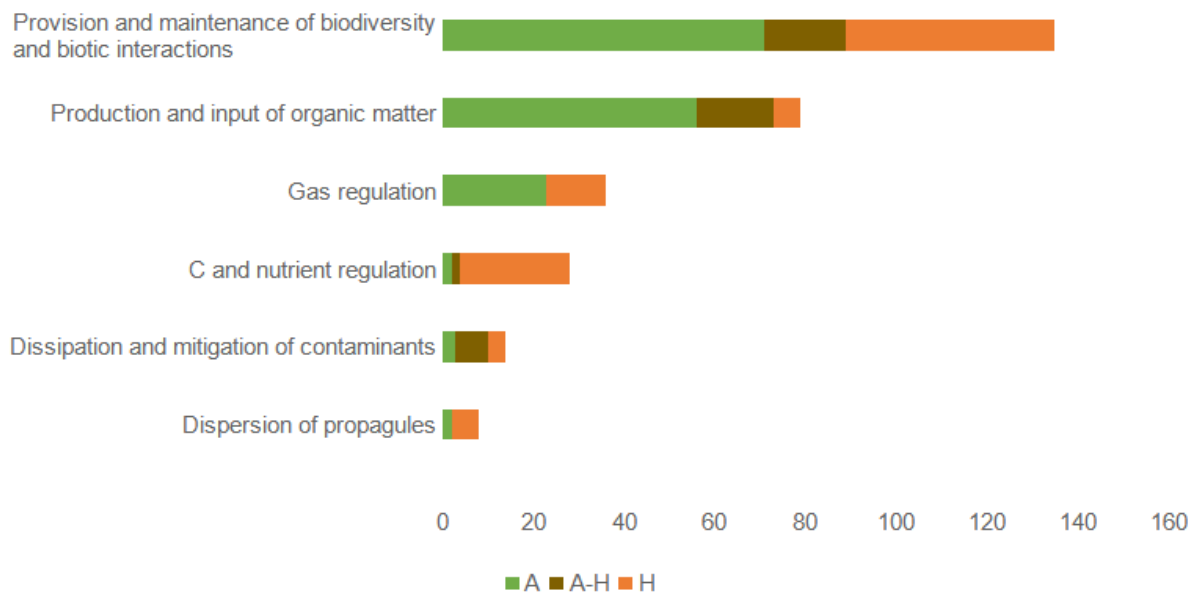
144 considering mixture effects. Autotrophic component (plot A) n = 108 occurrences, 45 chemicals.
145 Heterotrophic component (plot B) n = 120 occurrences, 34 chemicals.

146

147 *3.2. General overview of functional endpoints affected by pesticide exposure*

148 Aquatic microbial communities, including autotrophs and heterotrophs, support a large number of
149 ecosystem functions (Leenhardt et al. 2022) that can be impacted by pesticide exposure. While most
150 studies in the literature have addressed pesticide effects on the biomass and diversity of microbial
151 communities, only half attempted to assess pesticide effects on other ecosystem functions.
152 Moreover, a large number of publications mention functional endpoints as highly sensitive to
153 pesticide exposure, often providing earlier or exacerbated responses compared to structural
154 endpoints (such as taxonomic composition; e. g. Sabater et al. 2007).

155 The framework of clearly-defined core categories of ecosystem functions and services by Leenhardt
156 et al. (2022) helped us to identify which functions are effectively or potentially threatened by
157 pesticides (Table 2). Most studies on the heterotrophic component of aquatic microbial communities
158 have examined the effect of pesticides on functions related to the provision and maintenance of
159 biodiversity and biotic interactions (Figure 3). The effects of pesticides on heterotrophs' biodiversity
160 and interactions have been much more studied for bacteria than for fungal communities (65% and
161 33%, respectively), and the effects on protozoan communities remain almost unexplored (2%).
162 Besides, the processes of OM decomposition (36%) and respiration (14%) were the most examined
163 when assessing pesticide effects on carbon and nutrient cycles and gas regulation functions. To a
164 lesser extent, hyphomycete sporulation and bacterial production were employed to determine
165 pesticide effects on propagule dispersion and OM production, respectively. In the autotrophic
166 component, pesticides' impact on biodiversity was also widely studied (73%). Then, primary
167 productivity (58%) and photosynthesis (22%) impairment were the most examined processes when
168 assessing pesticide effects on the production of OM and gas regulation functions, which is consistent
169 with the large number of studies focusing on the impact of molecules with an herbicidal mode of
170 action (inhibitors of photosystems I and II, Figure 2). Few references explored the interactions with
171 higher trophic levels (8%), targeting biofilm grazers. Indeed, the large majority of the effects
172 assessed refer to the direct impacts of pesticides towards specific components of aquatic
173 communities, herbicides vs. autotrophic components and fungicides vs. heterotrophic components
174 (Figure 2). A limited number of articles report the indirect impact of pesticides in microbial
175 communities due to alterations in interactions between microorganisms (competition, facilitation; e.
176 g. Sura et al. 2012, Artigas et al. 2014) or with higher trophic levels (changes in grazing pressure; e. g.
177 Rohr & Crumrine 2005, Bundschuh et al. 2011, Neury-Ormanni et al. 2020).



179 Figure 3. Number of AME studies examining several ecosystem functions between 2000 and 2020 for
 180 autotrophic (A), heterotrophic (H), or both (A-H) components of aquatic microbial communities
 181 (n=168). Note that several functions may be considered in a single record.

182

183 The choice of the functional endpoints studied is generally adapted to the temporality of exposure to
 184 pesticides: physiological descriptors allow highlighting short-term toxicity, while impacts on biomass,
 185 or changes in taxonomic composition, generally operate for a longer duration of exposure (Sabater et
 186 al. 2007). Even if the choice of the experimental approach (microcosm, mesocosm, *in situ*; Table 1)
 187 has an influence on the results and conclusions that can be drawn, it did not determine the choice of
 188 functional parameters in the studies analysed here. For instance, molecular-level endpoints were
 189 used in field experiments (e. g. *nif* genes, Sun et al. 2012) and ecosystem-level functional endpoints
 190 were used in microcosm experiments (e. g. primary production, OM decomposition, Gardeström et al.
 191 2016). Nevertheless, experiments under controlled conditions (i. e. microcosms) tended to use
 192 molecular-level endpoints permitting a mechanistic understanding of pesticide effects on microbial
 193 processes. Finally, several studies used multiple endpoint approaches targeting the same microbial
 194 function (i. e. from the gene expression to the enzyme activity, e. g. Su et al. 2019). Despite various
 195 studies showing a clear correlation between responses of multiple endpoints to pesticides (e. g.
 196 Artigas et al. 2012b), others showed more mixed results (e. g. Pesce et al. 2016). For instance, the
 197 structural and functional endpoints of leaf-associated microbial assemblages can be decoupled when
 198 exposed to contaminants, suggesting that contaminants effects may be misinterpreted when solely
 199 based on structural endpoints (Feckler and Bundschuh 2020).

200 Table 2. Main ecosystem functions, processes and endpoints measured in microbial community ecotoxicology studies during between 2000 and 2020.
 201 Functions can be ensured by autotrophs (A), heterotrophs (H) or both (A and H, when one is dominant the other is mentioned in brackets). Examples of
 202 references (non-exhaustive) using the methods are provided, together with the main observed effects of pesticide exposure towards the corresponding
 203 ecosystem functions.

Ecosystem function	Ecosystem process	Main community targeted (A, H, A-H)	Endpoints used	Analyses and techniques	References using the method (examples)	Observed impact of pesticide exposure
Provision and maintenance of biodiversity and biotic interactions in aquatic ecosystems	Population/community dynamics	A-H	Growth rates	Cell increase over time using microscope counts	Moreira-Santos et al. (2005), Hayashi et al. (2011), Proia et al. (2011), Larras et al. (2016)	Decrease
				Cell increase over time using flow cytometry	de la Broise & Stachowski-Haberkorn (2012), Bricheux et al. (2013), Artigas et al. (2017)	
			Cell densities, live/dead ratio	Cell densities measurements using microscope counts	Perez et al. (2007), Debenest et al. (2009), Vera et al. (2010), Proia et al. (2011), Kim-Tiam et al. (2014, 2015), Lozano et al. (2018)	
	Biodiversity	A	Taxonomic composition	Taxonomic analysis using microscope identification	Bérard et al. (2003), Morin et al. (2009, 2010), Magnusson et al. (2012), Roubéix et al. (2012), Kim-Tiam et al. (2014, 2015), Feckler et al. (2018b), Smedbol et al. (2018), Bighiu et al. (2020)	Selection of pollution tolerant taxa/groups, to the detriment of sensitive taxa/groups
A	Pigment composition analysis using HPLC	Pinckney et al. (2002), Dorigo et al. (2007), Magnusson et al. (2012)				

		A		Pigment composition analysis using fluorimetry techniques	Morin et al. (2010), Kim-Tiam et al. (2015), Polst et al. (2018)	
		A-H		Community structure analysis using DGGE/TGGE	Dorigo et al. (2007, 2010), Tien et al. (2013), Paule et al. (2015a)	
		A-H		Community diversity analysis using high-throughput sequencing	Dimitrov et al. (2014), Lu et al. (2019)	
	Competition/facilitation	A-H	Diversity/co-occurrence analyses	Species density analyses using microscope counts	Proia et al. (2011)	Targeted groups outcompeted by non-target components
	Trophic interactions	A	Production of essential fatty acids	Analysis of fatty acid profiles using gas chromatography	Filimonova et al. (2016, 2018), Demailly et al. (2019), Zhang et al. (2020)	Decrease
		A-H	Trophic interactions through food web approaches	Multiple trophic levels design using density or diversity dynamics analyses	Rohr & Crumrine (2005), Chang et al. (2011), Cothran et al. (2011), Zubrod et al. (2011), Neury-Ormanni et al. (2020)	Weak indirect effects (herbicides), reduced top-down impacts (insecticides)
Production and input of organic matter in aquatic ecosystems	Primary productivity	A	Gross and net primary production	O ₂ production using respiration chambers	Murdoch et al. (2013)	Decrease (herbicides), no effect or increase (fungicides, insecticides)
				Inorganic ¹⁴ C incorporation using radioisotopic techniques	Vera et al. (2010), Tlili et al. (2011), Villeneuve et al. (2011)	
			Chlorophyll-a concentration	Chlorophyll-a concentration analysis using HPLC	Gustavson et al. (2003), Magnusson et al. (2012), Feckler et al. (2018b), Bighiu et al. (2020)	

				Chlorophyll-a concentration analysis using spectrophotometric techniques	Morin et al. (2010), Murdoch et al. (2013), Abhelo et al. (2016), King et al. (2016), Lozano et al. (2018), Smedbol et al. (2018)	
				Chlorophyll-a concentration analysis using fluorimetry techniques	Bérard et al. (2003), Schmitt-Jansen & Altenburger (2005), Pesce et al. (2010)	
	Biomass production	A-H	Periphyton production over time	Determination of Dry weight, Ash-Free Dry Mass, Particulate C, over time	Dorigo et al. (2010), King et al. (2016), Feckler et al. (2018b)	Decrease at elevated concentrations of exposure
		H	Fungal production over time	Analysis of ergosterol concentration over time using HPLC	Artigas et al. (2012), Dimitrov et al. (2014), Gardeström et al. (2016)	Decrease in fungal production (fungicides) and increase in bacterial production
		H	Bacterial production over time	Leucine incorporation over time using radioisotopic techniques	Widenfalk et al. (2004), Widenfalk et al. (2008), Milenkovski et al. (2010)	
				Thymidine incorporation over time using radioisotopic techniques	Pesce et al. (2006), Pesce et al. (2008), Sura et al. (2012), Artigas et al. (2014)	
Gas regulation	C cycle: Photosynthesis	A	Photosynthetic activity	Inorganic ¹⁴ C assimilation using radioisotopic techniques	Gustavson et al. (2003), Schaffer & Sebetich (2004), Perez et al. (2007)	Decrease (herbicides), sometimes recovering over

			Photosystem II efficiency using PAM techniques	Schmitt-Jansen & Altenburger (2005), Ricart et al. (2009), Magnusson et al. (2012, 2013), Smedbol et al. (2018)	time after low-dose exposure
C cycle: Respiration	H (A)	Dissolved oxygen consumption rate	Oxygen consumption analysis using respiration chambers	Kreutzweiser et al. (2007)	Decrease (fungicides), neutral or stimulatory effects of herbicides and insecticides
			CO2 production analysis using 5-Cyano-2,3-ditoyl tetrazolium chloride (CTC) reduction	Pesce et al. (2018), Pesce et al. (2006)	
		Carbon dioxide production rate	CO2 production analysis using Gas chromatography, Substrate-induced respiration (MicroResp)	Chen et al. (2019), Artigas et al. (2014), Mbanaso et al. (2014), Widenfalk et al. (2004)	
N cycle: Denitrification	H	N fluxes	Denitrification flux analysis using the acetylene inhibition method	Milenkovski et al. (2010)	No effect or negative for fungicides
			N flux analysis using 15N isotopic techniques	Widenfalk et al. (2004), Su et al. (2019), Chen et al (2019)	

			Denitrification-involved enzymes rates (nitrate reductase, nitrite reductase, nitric oxide reductase, nitrous oxide reductase)	Enzyme activity measurements using dyes (ex. Viologen) reduction bioassay	Su et al. (2019), Chen et al (2019)	
	S cycle: Sulfate reduction	H	Sulfate reduction rate	Sulfate reduction analyses using turbidimetric method (sulfate) and copper spectrophotometric method (sulfide)	Garcia-Cruz et al. (2010)	Decrease (herbicides and metabolites)
Carbon and nutrient regulation in aquatic ecosystems	Organic carbon and nutrient recycling	A-H	N-uptake (NO ₃)	N uptake analysis using Tracer addition techniques	Mulholland et al. (2004)	Transient decay then recovery
			N-uptake (NH ₄)	N uptake analysis using ammonium decay measurements	Murdoch et al. (2013)	
			P-uptake	P uptake analysis using soluble reactive phosphorus measurements	Proia et al. (2011)	
		H	Decomposition rates	Analysis of mass loss of leaf litter, cotton or wood strips	Artigas et al. (2012), Dimitrov et al. (2014), Kreuzweiser et al. (2007), Brosted et al. (2016), Rossi et al. (2018)	decrease (or no effect)

			Extracellular enzymatic activity rates (cellulolytic, ligninolytic, peptidases, phosphatases)	Enzyme activity measurements using fluorescent methylumbelliferone-substrate analogues	Ricart et al. (2009), Artigas et al. (2012b), Donnadiu et al. (2016), Rossi et al. (2018)	variable effect depending on pesticide and enzyme activity type
			Metabolic richness and diversity	C-substrates utilisation using Biolog Ecoplates	Campbell et al. (2003), Foley et al. (2008), Sura et al. (2012), Paule et al. (2015a,b), Pringault et al. (2016), Lv et al. (2017), Kumar et al. (2020)	
Dissipation and mitigation of contaminants	Transformation and mineralisation of contaminants	H (A)	Dissipation rates	Contaminants dissipation analysis using mass balance calculations	Thomas & Hand (2011), Tien et al. (2013), Paule et al. (2015a), Carles et al. (2017)	Increase
		H	Mineralization rates	Contaminantes mineralisation analysis using radiorespirometry techniques	Pesce et al (2013), Pesce et al. (2010)	
	Removal of contaminants	A-H	Organic pesticide bioaccumulation	Contaminants accumulation analysis using GC-MS analysis	Lawrence et al. (2001), Zhao et al. (2015), Qiu et al. (2017), Rooney et al. (2020)	
		HPLC-MS, UPLC-ToF analysis		Chaumet et al. (2019)		
Dispersion of propagules in aquatic ecosystems	Sporulation	H	Hyphomycetes sporulation rates	Sporulation rates determination using conidia counts and identification under the microscope	Flores et al. (2014), Donnadiu et al. (2016), Brosed et al. (2016), Pimentao et al. (2020)	Decrease

206

208

209 3.3. Impacts of pesticides on ecosystem functions ensured by autotroph-dominated microbial
210 communities

211

212 *Provision and maintenance of biodiversity*

213 In the autotrophic component of microbial communities, the impairment of biodiversity by
214 herbicides is one of the impacts most frequently studied, thanks to a large range of available
215 methods. Classic microscopic methods allowing algal identification were often complemented by
216 other techniques, from rough estimates of algal groups based on pigment profiles to genetic
217 approaches (electrophoresis being recently supplanted by high-throughput sequencing). Several studies
218 demonstrated changes in the algal composition and diversity of phytoplankton and periphyton
219 exposed to herbicides, exposure generally leading to the selection of pollution-tolerant algal groups
220 to the detriment of sensitive individuals. In the presence of the broad-spectrum herbicide
221 glyphosate, studies observed higher vulnerability of chlorophytes and diatoms to herbicides, and
222 increasing amounts of cyanobacteria (Pérez et al. 2007, Smebold et al. 2018). More contrasted
223 responses of algal groups (diatoms in particular) were shown with other herbicides, such as the
224 widely studied photosynthesis inhibitor diuron. Community changes induced by diuron as well as
225 other pesticides were assessed by Dorigo et al. (2007, 2009, 2010) in the Morcille watershed (South-
226 East France). Their work highlighted the increase of diatoms and cyanobacteria to the detriment of
227 green algae along the contamination gradient, based on pigment signatures (HPLC measurements).
228 Villeneuve et al. (2011) assessed the effects of long-term exposure of biofilms to diuron and the
229 fungicide azoxystrobin by microscopic counts; they found that diatoms were more tolerant to
230 pollution than cyanophytes and chlorophytes. Using fluorescence measurements, on the contrary,
231 higher amounts of green algae were detected downstream of pesticide inputs (Kim-Tiam et al. 2014)
232 in the same watershed, reflecting interannual variability or divergences between microscopy, HPLC
233 and fluorimetry results. The application of the herbicide simetryn on phytoplankton communities
234 caused a decrease in the abundance of chlorophyceae (*Chlamydomonas* sp.) but did not affect
235 significantly diatoms (Chang et al. 2011), confirming the ability of the latter to maintain under
236 exposure to photosynthesis inhibitors.

237 Note that the vast majority of the effects assessed refer to direct impacts of herbicides on
238 community composition; however, some studies report indirect effects of pesticides, related to
239 changes in the relationships between organisms in the community. An overall decrease in
240 biodiversity was also demonstrated with fungicides. For instance, Lu et al. (2019) exposed
241 phytoplankton to azoxystrobin; they concluded that competition relationships were altered under
242 pesticide exposure, favouring cyanobacterial dominance at the detriment of chlorophytes. Abelho et
243 al. (2016) found that the fungicide pyrimethanil decreased fungal biomass in biofilms, altering the
244 relative abundances of periphytic fungi and algae. On the contrary, exposure to the insecticide
245 pyridaben enhanced the diversity of planktonic algal communities (Rand et al. 2001), compared to
246 reference conditions. This could be due to decreased top-down control or medium enrichment by
247 dead insect bodies (Knapp et al. 2005).

248 The impacts of pesticide exposure on biodiversity considering species-specific sensitivities of the
249 autotrophic component of microbial communities mostly targeted diatoms, probably as a
250 consequence of their tolerance to various pesticides (see above, or Rimet & Bouchez 2011; Bricheux
251 et al. 2013) and of their wide use as environmental bioindicators (Coste et al. 2009; Morin et al.

252 2016; Water Framework Directive 2000/60/EC). As for algal groups, a selection of tolerant species
253 was often observed, together with a general decrease in species diversity. In several field studies
254 attempting to characterize the specific sensitivity of taxa to pesticides, diatom diversity metrics
255 highlighted eutrophication more than pesticide exposure (Morin et al. 2009, 2010; Roubeix et al.
256 2012; Wood et al. 2019; Bighiu et al. 2020), as both pollutions often occur simultaneously in
257 agricultural watersheds. This may explain why diatom species able to colonize
258 eutrophic/hypereutrophic environments, such as *Nitzschia palea* or *Planothidium frequentissimum*,
259 were considered more tolerant to herbicides (e. g. Debenest et al. 2010). To disentangle the nutrient
260 from the pesticide effects, works were performed under laboratory-controlled conditions (e. g.
261 Villeneuve et al. 2011; Tien et al. 2013; Bayona et al. 2014; Wood et al. 2017) and generally
262 highlighted a pesticide-induced selection of tolerant taxa and an overall decrease in diversity. Other
263 confounding factors were found to mask the deleterious impacts of pesticides on the taxonomic
264 structure of periphyton, such as light availability (Feckler et al. 2018b). Another approach combining
265 diatom traits, taxonomic metrics and diatomic indices in multimetric models was proposed by Larras
266 et al. (2017). They showed that pesticides selected for motile diatom species, highlighting the
267 promising use of approaches based on traits or ecological life forms (Rimet & Bouchez 2011, Marcel
268 et al. 2013) for assessing toxic-induced species selection in diatoms.

269

270 *Provision and maintenance of biotic interactions*

271 As primary producers, photosynthetic microorganisms are essentially at the base of aquatic food
272 webs. The cascading effects of pesticides along the trophic chain have been examined mainly under
273 controlled conditions.

274 Direct deleterious effects of herbicides on the autotrophic component of microbial communities
275 were sometimes shown to decrease the biomass of grazers, with effects that are difficult to detect in
276 most cases. For instance, Rohr & Crumrine (2005) demonstrated that the decrease in periphyton
277 biomass after atrazine exposure caused a decrease in the biomass and reproduction of snails and
278 tadpoles. In contrast, Muñoz et al. (2001) found, in a simple periphyton-snails design, that the
279 biomass of periphyton decreased with the addition of atrazine, while that of grazers was not affected
280 by the herbicide. Chang et al. (2011) assessed how another triazine herbicide, simetryn, impacted a
281 planktonic microbial food web composed of phytoplankton, bacteria, heterotrophic nanoflagellates,
282 and ciliates. The highest concentration caused a decrease in the biomass of phytoplankton, especially
283 in green algae, but weak direct and indirect effects on zooplankton. They suggested that other
284 microorganisms unimpacted by the herbicide could constitute alternative food for zooplankton. In
285 complement, Neury-Ormanni et al. (2020) suggested that a poor microalgae quality subsequent to
286 diuron exposure (e. g. decreased content in essential fatty acids, see below), could trigger
287 invertebrates to graze higher amounts of contaminated microalgae, to compensate for the decrease
288 in nutrient supply (also see “Dissipation and mitigation of contaminants”). The impacts of glyphosate
289 on phytoplankton were also shown to be altered in the presence of golden mussels (*Limnoperna*
290 *fortunei*), which addition modified nutrient content in the water column and selected for
291 phytoplankton species (De Stefano et al. 2018; Gattás et al. 2018). Such results contrast those of
292 Iummato et al. (2017) who found no differences in periphyton exposed or not to glyphosate in the
293 presence of *L. fortunei* suggesting that the mussels attenuated the effects of glyphosate by
294 contributing to its dissipation, or of López-Doval et al. (2010) who did not observe any significant
295 interactions between diuron, biofilms and grazers (freshwater snails *Physella acuta*).

296 On the other side, the exposure of herbivorous grazers to insecticides can modulate the responses of
297 phototrophic communities: the interactions are often marginal but can be notable in the case of
298 insecticides (Cothran et al. 2011; Hua & Relyea 2012; Neury-Ormanni et al. 2020) where there are
299 few direct effects on primary producers but reduced top-down pressure resulting from toxicity on
300 grazers, indirectly favouring the development of algae.

301 At the molecular level, some studies looked at the synthesis of proteins, fatty acids or carbohydrates,
302 and the incorporation of nutrients (cited for example in Debenest et al. 2010). Such biomolecules are
303 key in the diet of higher organisms and the provision of energy to the consumers; the impairment of
304 the synthesis of fatty acids is recently given increasing attention with the development of targeted
305 and non-targeted metabolomics (Zhang et al. 2020). The supply of essential fatty acids (omega 3 and
306 omega 6) along the trophic chain is mainly ensured by microalgae, in particular diatoms (Gonçalves
307 et al. 2016). The consequences of pesticide exposure on their nutritional value are crucial for
308 ecosystem functioning, as the impacts on the basis of the food web could have severe repercussions
309 for higher trophic levels. Herbicide-induced alterations in fatty acid profiles were noticeable in
310 decreasing the proportion of highly unsaturated fatty acids including the eicosapentaenoic acid
311 (omega 3) of monospecific cultures of diatoms (Filimonova et al. 2016, 2018; Demailly et al. 2019).
312 Conversely, in a metabolomic study, Zhang et al. (2020) observed a significant upregulation of some
313 omega 3 and omega 6 fatty acids (e. g. linolenic acid, linoleic acid and arachidonic acid) in a
314 phytoplanktonic community exposed to the fungicide azoxystrobin, likely a consequence of reduced
315 competition in the community.

316 Finally, certain photosynthetic organisms can be sources of toxic molecules, and the presence of
317 pesticides may favour them directly. For example, Lürling & Roessink (2006) demonstrated that the
318 cyanobacteria *Microcystis aeruginosa* outcompeted chlorophytes (*Scenedesmus obliquus*) in the
319 presence of the herbicide metribuzin. In the same way, Khadra et al. (2018) showed that glyphosate
320 exposure was likely to induce blooms of the potentially toxic genus *Anabaena* in lacustrine biofilms.

321 Although many authors mentioned the impairment of food provision/energy supply as a possible
322 consequence of the toxic effects measured, those studies rarely addressed directly the impact of
323 pesticide exposure on the transfer of OM and energy to higher trophic levels.

324

325 *Production and supply of organic matter*

326 To assess the impacts of herbicides, autotrophic biomass is often mentioned in freshwater aquatic
327 microbial communities, together with photosynthesis (see “Gas regulation”). Literature reviews
328 (Pesce et al. 2011, or Debenest et al. 2010 focusing on diatoms) highlighted the negative effects of
329 herbicides on the algal component of freshwater biofilms, using biomass, microscope countings, or
330 pigment content. Besides rough estimates such as dry weight or ash-free dry mass, concentrations of
331 chlorophyll-*a* are often used as an estimate of the biomass of photosynthetic microorganisms in
332 phytoplankton and periphyton. Considering freshwater microalgae, the recent review by Vonk &
333 Kraak (2020) confirmed that a high number of studies dealt with the ecotoxicity of pesticides
334 targeting photosynthesis (triazines, ureas) and other herbicides (e. g. chloroacetamides) based on
335 global endpoints such as abundance, biomass or growth descriptors. For most of these compounds,
336 median EC_{50s} was below the mg/L range, suggesting that toxicity should occur at environmental
337 concentrations with visible impacts on OM production by autotrophs.

338 In most studies, long-term exposure (weeks to months) to herbicides impaired the function of
339 production and supply of OM by autotrophic components of microbial communities. These
340 herbicides reduced the chlorophyll-*a* concentration (Murdoch et al. 2013; Rohr & Crumrine 2005),
341 growth dynamics and primary production (measured as ¹⁴C incorporation) of periphytic
342 communities (Villeneuve et al. 2011). In several field studies where exposure to pesticides is complex
343 (mixtures of molecules) at concentrations in the range of ng-µg/L, negative impacts on periphytic
344 biomass were observed (e. g. Morin et al. 2010; Kim-Tiam et al. 2014). The diversity of works
345 assessing the growth responses of periphyton exposed to one particular pesticide shows that
346 exposure concentration and the chemical composition of pesticides matter. Dealing with glyphosate
347 for instance, Smedbol et al. (2018) used environmentally realistic concentrations of the herbicide
348 (from µg to mg of active ingredient (a.i.)/L) and demonstrated a decrease in chlorophyll-*a* and
349 carotenoid concentrations of phytoplankton communities from the lowest concentration of
350 exposure. Opposite results were found by Pérez et al. (2007) using higher concentrations of
351 glyphosate: the primary production of phytoplankton exposed to milligrams a.i./L of glyphosate
352 roughly doubled compared to control conditions, which could be related to increased phosphorus
353 concentrations in the medium supplied by glyphosate additions (Saxton et al. 2011, but see Carles &
354 Artigas 2020). Such an increase in biomass (assessed as dry weight or chlorophyll-*a* concentration)
355 was confirmed recently by Vera & Trinelli (2021) after a 7-day exposure of periphyton to
356 concentrations around the mg a.i./L of glyphosate. In other cases, glyphosate exposure had no effect
357 on chlorophyll *a* content but only impacted their structural composition (see “Provision and
358 maintenance of biodiversity”), highlighting the capacity of biofilms to regulate and maintain a
359 constant level of algal biomass (Pesce et al. 2009; Bricheux et al. 2013).

360 Contrastingly, studies using pesticides other than herbicides generally did not observe a decrease in
361 microalgal biomass (e. g. Artigas et al. 2014, for the fungicide tebuconazole), sometimes even an
362 increase. For instance, Abhelo et al. (2016) found increased microalgal biomass compared to fungal
363 biomass, under exposure to the fungicide pyrimethanil. Rohr & Crumrine (2005) found that exposure
364 to the insecticide endosulfan did not affect the biomass of periphytic algae, estimated by chlorophyll-*a*.
365 Rand et al. (2001) showed no effects on periphyton communities but highlighted a positive
366 correlation of phytoplankton abundance with exposure to the insecticide-miticide pyridaben. In such
367 studies, it is important to highlight that a “positive” impact of pesticides on growth may rather derive
368 from decreased competition between microbial components or reduced grazing (i. e. top-down
369 pressure) (Rand et al. 2001), as described in “Provision and maintenance of biotic interactions”.

370 Albeit the effects of pesticides on this function are widely studied, most of these works have
371 estimated effects via proxies under controlled conditions rather than measuring effects in the field.

372

373 *Gas regulation*

374 Some studies also looked at the consequences of pesticide exposure on gross primary production
375 and respiration in autotrophic biofilms, generally by recording dissolved oxygen in incubation
376 chambers, under light and dark conditions respectively. Respiration was, in general, unimpacted by
377 pesticide exposure, while some studies highlighted a decrease in gross primary production after
378 contamination. Murdoch et al. (2013) measured these functions in wetland periphyton after an
379 agricultural runoff. They found interactive effects with total phosphorus causing a respiration
380 decrease, and atrazine mitigating the negative relationship between respiration and phosphorus.

381 Using laboratory chambers, they found no impact of atrazine on the respiration of periphyton on the
382 third day of exposure but a decrease in gross primary production at the highest concentration of
383 exposure. Villeneuve et al. (2011) also observed a decrease in the primary production of biofilms
384 exposed to a mixture of pesticides, even if no decrease was detected in algal density. Lozano et al.
385 (2018) found transient effects of herbicides (2,4-D, glyphosate and their mixture) on the respiration
386 of freshwater phytoplankton. Immediately following application, the herbicides caused a decrease in
387 the respiration rate of microalgal communities in mesocosms, but after 1 day the effect was no
388 longer detected. This may explain why, in river biofilms exposed to glyphosate, Artigas et al. (2020)
389 found no significant impact on net primary production nor respiration rates analysed from 4 days and
390 later on.

391 Despite the photosynthetic character of microalgae, the influence of pesticides on gas regulation in
392 microalgal communities remains poorly studied in freshwaters. The general decline in research
393 dedicated to primary productivity in periphyton over the last three decades was recently highlighted
394 by Zhao et al. (2022). At a global scale, the part of freshwaters in the total amount of water on planet
395 Earth is very low (1%) compared to oceans (> 95%) where the contribution of phytoplankton to the
396 world's primary production is paramount (Field et al. 1998). Moreover, with the development of
397 PAM (Pulse Amplitude Modulated) fluorimetry techniques over the last 20 years, chlorophyll-*a*
398 fluorescence parameter (e. g. photosynthetic efficiency) are increasingly used as endpoints of
399 pesticide toxicity (Table 2; this proxy representing 76% of photosynthesis assessments in the corpus).
400 Chlorophyll-*a* fluorescence bioassays have been applied successfully to assess the ecotoxicity of a
401 wide range of contaminants (e. g. metals, herbicides, petrochemicals), as such tests are rapid, non-
402 invasive and non-destructive (Ralph et al. 2007). Most of the studies retrieved using PAM techniques
403 highlighted photosynthesis impairment with herbicide exposure; however, such measurements stray
404 from the primary assessment of the ecosystem function of gas regulation related to photosynthesis.

405

406 *Nutrient regulation*

407 The impacts of pesticides on the function of regulation of nutrient cycles relative to freshwater
408 microalgae are mentioned, for example in the review by Debenest et al. (2010). Some herbicides can
409 reduce the absorption of nutrients (nitrates, nitrites, phosphorus and silica, in particular). In their
410 review of the environmental consequences of herbicide impacts on cyanobacteria, Brêda-Alves et al.
411 (2021) showed that increasing concentrations of several herbicides in aquatic environments impair
412 atmospheric nitrogen fixation by cyanobacteria, as a result of photosynthesis inhibition. Such a
413 decrease is likely to impact the overall aquatic nitrogen cycle. However, in their study addressing the
414 effects of atrazine on laboratory periphyton, Murdoch et al. (2013) observed an early, sharp
415 reduction of NH₄⁺ uptake rates (75 to 84%) after 3 days of exposure, followed by a recovery of
416 control rates after 1 week. According to these observations, the effects of pesticide exposure on
417 nutrient cycles may be transient. Concerning P cycle, Proia et al. (2011) did not observe any change in
418 phosphorus uptake by periphyton exposed for 2 weeks to diuron.

419

420 *Dissipation and mitigation of contaminants*

421 Living in biofilms can buffer microorganisms from variations in the external environment, including
422 exposure to pesticides. Besides, autotrophic biofilms can bioaccumulate, and thus partly remove,
423 organic pesticides from water. In their recent review, Bonnineau et al. (2021) demonstrated that

424 freshwater periphyton can accumulate organic pesticides present in the water column with bio-
425 uptake efficiencies varying according to their hydrophobicity, estimated by their partition coefficient
426 between octanol and water (log K_{ow}). Besides, Vonk & Kraak (2020) reported that uptake rates are
427 influenced by biological characteristics (cell size and lipid composition of the organisms).
428 Bioconcentration factors are calculated as the ratio of pesticide concentration in the biofilm with
429 respect to its concentration in surrounding water, and values over 1 indicate bioconcentration. In
430 Canadian wetlands (Rondeau Bay, Ontario), Rooney et al. (2020) found that periphyton
431 bioconcentrates a large variety of pesticides, with bioconcentration factors ranging from 12 for the
432 herbicide dicamba up to 6864 for the fungicide boscalid. Removal of pesticides by microalgae from
433 their environment was also shown to occur in phytoplankton. In lakes, high bioconcentration factors
434 for organochlorine pesticides were detected in phytoplankton (Zhao et al. 2015; Qiu et al. 2017), with
435 a high affinity of pesticides for diatoms and cryptophytes. However, bioconcentration of pesticides in
436 primary producers may expose other aquatic biota via consumption (Qiu et al. 2017; Rooney et al.
437 2020), and therefore have deleterious consequences on biotic interactions (“Provision and
438 maintenance of biotic interactions”).

439 The capacity of microalgae to detoxify organic contaminants has been shown using several
440 wastewater-treatment microalgal technologies (e. g. Pazos et al. 2016; Sutherland & Ralph 2019);
441 data in natural aquatic ecosystems are scarce. Paule et al. (2015) compared the removal rates of
442 alachlor by wastewater and river biofilms showing that, even 10 times lower for natural biofilms, the
443 rates of disappearance of the pesticides reached 5-10 µg alachlor removed daily per gram of biofilm
444 dry weight. Such transformation capacities of natural periphytic biofilms can contribute to the self-
445 purification of rivers downstream pesticide inputs. In their review on cyanoremediation, Kumar and
446 Singh (2017) highlighted the biodegradation capabilities of cyanobacteria, with some genera
447 common in freshwaters (e. g. *Anabaena*, *Microcystis*, *Nostoc*, *Spirulina*) able to degrade various
448 pesticides. Tien et al. (2013) assessed the capacity of freshwater autotrophic biofilms to biodegrade
449 carbamate pesticides (methomyl, carbaryl, carbofuran). Although toxic effects of the pesticides were
450 observed on communities of diatoms and bacteria (See “Provision and maintenance of biodiversity”),
451 they found that tolerant diatoms and bacteria were potential degraders of the three carbamate
452 pesticides tested alone. However, their ability to break down the pesticides was impaired when
453 mixtures were tested, suggesting a decrease in rivers' bioremediation ability under complex
454 contamination conditions. In another study, Lawrence et al. (2001) proved that atrazine and diclofop
455 methyl were mineralized by river biofilms to CO₂. In most studies, observed biotransformation
456 reactions mostly corresponded to substitution-type reactions catalyzed by central metabolic
457 enzymes ubiquitously found in bacteria. Although correlations with any autotrophic component were
458 not assessed, similar enzymatic machinery exists in microalgae (Sheng et al. 2022). Thomas & Hand
459 (2011) showed that, in environmental systems, the presence of algae increased the rate of
460 degradation of several pesticides compared to sediment alone.

461

462 Summarizing the main data available, pesticide exposure often impacted the following ecosystem
463 functions supported by autotrophs in microbial communities:

464 -Provision and maintenance of biodiversity and trophic interactions, highlighting the selection of
465 tolerant microalgae (taxa or groups) and a decrease in diversity, while cascading effects were

466 generally weak or hardly observable. However, the current development of dietary tracers (e. g. fatty
467 acids) is a promising approach to shape future research;

468 -Production and supply of OM, showing a general trend of decreasing microalgal biomass with
469 herbicide exposure, while fungicides and insecticides did not significantly impair autotrophic biomass
470 as the result of decreased competition or predation;

471 -Gas regulation, through the reduction of gross primary production and impairment of
472 photosynthesis.

473

474 *3.4. Impacts of pesticides on ecosystem functions ensured by heterotroph-dominated microbial*
475 *communities*

476 *Provision and maintenance of biodiversity and biotic interactions*

477 Microbial heterotrophs play a pivotal role in aquatic food webs as both organic matter (OM)
478 decomposers and OM suppliers to higher trophic levels (Gessner and Chauvet 1994). Little is known
479 about cascading effects of pesticides exposure on aquatic food webs, especially for the heterotrophic
480 component of microbial communities (e. g. Zubrod et al. 2011). Most of these experiments were
481 conducted in microcosms and revealed both “top-down” and “bottom-up” effects in natural
482 microbial communities resulting from pesticide exposure. For instance, exposure to low deltamethrin
483 concentrations increased drastically arthropod mortality followed by a sudden increase of activity in
484 bacterial and algal communities in the water column (Knapp et al. 2005). This sequential response
485 can be explained by an apparent sudden release of nutrients following the death of the arthropods,
486 which triggered a series of responses in the microbial loop. Pesticides can also reduce OM
487 consumption by several aquatic invertebrate species (e. g. Zubrod et al. 2015), often driven by
488 changes in the OM-associated microbial community (e. g. Feckler et al. 2016). For instance, the
489 exposure to a mixture of five current-use fungicides (azoxystrobin, carbendazim, cyprodinil,
490 quinoxyfen, tebuconazole) reduced leaf consumption by gammarids, probably due to the reduction
491 in the richness of fungal species (ca. 40%) which contribute to reducing the nutritional quality of the
492 leaves (Zubrod et al. 2015). Diet-related effects impairing the functioning of the shredder *Asellus*
493 *aquaticus* resulting from epoxiconazole exposure were partially explained by lowered microbial
494 biomasses and altered composition of fatty acids associated with the leaf material (Feckler et al.
495 2016).

496 Few studies have attempted to investigate the effect of pesticides in microbial heterotrophs
497 interactions (i. e. fungus-fungus, bacteria-fungi, bacteria-protzoa, etc...) in the aquatic environment
498 (see review by Proia et al. 2012). For instance, the fungicide tebuconazole significantly reduced the
499 fungal biomass and increased that of bacteria in leaves and stream sediments in a microcosm
500 experiment (Donnadieu et al. 2016). The authors suggested that the increase in bacterial biomass
501 could be explained by (i) reduced resource competition between fungi and bacteria when fungi are
502 stressed by the fungicide and/or (ii) enhanced supply of nutrients released by the killed fungi and
503 further used by bacteria to grow. Indeed, this second hypothesis was also observed in soil microbial
504 communities after tebuconazole application (Cycoń et al. 2006). Within a biofilm, the effects of
505 grazing by protozoa on the structure of the bacterial community have been demonstrated (e. g. Matz
506 and Kjelleberg 2005). In this sense, the study of Friberg-Jensen et al. (2003) shows that reduced

507 grazer control from crustaceans due to cypermethrin exposure resulted in the proliferation of
508 rotifers, protozoans, bacteria, and algae from plankton and periphyton.
509

510 *Production and supply of organic matter*

511 Few studies have investigated the effect of pesticides on OM production by aquatic microbial
512 heterotrophs (e. g. Artigas et al. 2014, Pesce et al. 2006). In these studies, bacterial production was
513 analysed with two different radioisotopic techniques: leucine and thymidine incorporation. The
514 effect of herbicides alone or in mixtures, as well as fungicides alone or in mixtures, were tested on
515 bacterial production in periphytic, planktonic and sediment microbial communities. The literature
516 review highlighted that pesticides have little impact on bacterial production, with the vast majority of
517 responses being neutral or transient.

518 For instance, bacterial production measured in plankton and sediment microbial communities from
519 European lakes showed a transient decrease following exposure to the fungicides tebuconazole
520 (Artigas et al. 2014) and captan (Widenfalk et al. 2004), as well as exposure to the insecticides
521 deltamethrin and pirimicarb (Widenfalk et al. 2004). Exposure to environmentally realistic
522 concentrations of the herbicides diuron and glyphosate did not influence bacterial production in
523 stream periphyton communities (Pesce et al. 2006; 2009).

524 Studies in microcosms made it possible to assess the impact of pesticide cocktails on bacterial
525 production in aquatic microbial communities. Overall, no effects of cocktails of fungicides
526 (Milenkovski et al. 2010), herbicides (Sura et al. 2015; Pringault et al. 2016) or mixtures of herbicides,
527 fungicides and insecticides molecules (Widenfalk et al. 2008) were observed on bacterial production.
528 Field studies concluded also that pesticide contamination gradients observed in rivers and wetlands
529 from Europe weakly affected the bacterial production in aquatic microbial communities (Pesce et al.
530 2008 and Sura et al. 2012, respectively).

531 Fungal production measured as acetate incorporation into ergosterol or biomass accrual estimates
532 has not been employed in aquatic microbial ecotoxicology studies during the last 20 years. Instead,
533 fungal production in the form of conidia (sporulation) has been widely used as an endpoint to assess
534 pesticide effects on aquatic hyphomycetes communities. We decided to address pesticide effects on
535 aquatic hyphomycetes sporulation in the section below “Dispersion of propagules”.

536

537 *Gas regulation*

538 The assessment of the impact of pesticides on microbial gas exchanges with the atmosphere has
539 focused mainly on the carbon and nitrogen cycles (e. g. Widenfalk et al. 2004, Chen et al. 2019).
540 Studies assessing the impact of pesticides on the sulfur- and methane gases are rare in literature (e. g.
541 Garcia-Cruz et al. 2010). The study of endpoints related to respiration and denitrification processes is
542 the most used in aquatic microbial ecotoxicology. Respiration and denitrification appeared less
543 responsive to pesticides in aquatic environments with an equal number of studies showing negative
544 or neutral effects. Respiration rates in sediment microbial communities from a reservoir system in
545 China decreased as a function of a gradient of fungicide (chlorothalonil) and insecticide
546 (propramphos) concentrations (Su et al. 2019, Chen et al. 2019). Similar results were observed in
547 sediment microbial communities from an estuary zone in England (Garcia-Ortega et al. 2011). The
548 exposure history of microbial communities to pesticide contamination appears to determine the
549 response of respiration to the fungicide tebuconazole (Artigas et al. 2014). For instance, periphyton

550 respiration rates decreased after exposure to environmental concentrations of tebuconazole in a site
551 less contaminated by pesticides than in a site more contaminated by pesticides. Substrate-induced
552 respiration (SIR) is one of the most frequently used techniques to estimate respiration rates in
553 microbial communities (e. g. Tlili et al. 2011). It is important to note that SIR measurements include
554 the contribution of both autotrophs and heterotrophs in the respiration process, and therefore
555 caution must be taken when distinguishing pesticide effects between components of the microbial
556 community. The accumulation of high concentrations (mg/L) of metabolites from the herbicide 2,4-D
557 (4-chlorophenol, 2-chlorophenol and phenol) can inhibit the respiration rates from biofilms
558 dominated by sulfur-reducing bacteria (Garcia-Cruz et al. 2010). High concentrations of the herbicide
559 glyphosate can display neutral or stimulating effects on the respiration rates of microbial
560 communities from streams (Artigas et al. 2020) and ditch systems (Mbanaso et al. 2014), respectively.
561 Neutral effects of the herbicide diuron and the insecticide imidacloprid were observed in
562 communities from epilithon (Pesce et al. 2006) and associated with decomposing leaf litter
563 (Kreutzweiser et al. 2007) in streams, respectively. Laboratory studies (Widenfalk et al. 2004) and
564 field studies (Pesce et al. 2008) have coincided in describing the neutral effects of pesticide cocktails
565 (composed of fungicides, herbicides and insecticides) on the respiration of heterotrophic microbial
566 communities, as previously mentioned for autotrophs (see 3.3).

567 The denitrification rates and enzymatic activities involved in the denitrification activity in sediments
568 from the Three Gorges reservoir (China) decreased according to the increase in chlorothalonil
569 concentrations (Chen et al. 2019; Su et al. 2019). However, the expression of genes involved in the
570 denitrification activity (e. g. *nirK*, *nirS*, *narG*...) were not sensitive to chlorothalonil. These results from
571 these experiments show that multi-marker approaches (gene expression versus enzyme activity
572 measurement) often used in aquatic microbial ecotoxicology may result in contradictory conclusions.

573

574 *Carbon and nutrient regulation*

575 Pesticides impact has been extensively studied in the decomposition process of particulate OM
576 (mostly leaf litter) by heterotrophic components of microbial communities. This process has been
577 mostly examined in stream and river ecosystems where the availability of allochthonous OM prevails
578 over that of autochthonous origin (e. g. Zubrod et al. 2011, Rasmussen et al. 2012, Rossi et al. 2018).
579 Two main functional endpoints are used to assess pesticide effects on OM decomposition: the mass
580 loss of plant materials (e. g. Brosted et al. 2016) or the measurement of extracellular enzymatic
581 activities involved in the decomposition of plant materials (e. g. Artigas et al. 2012b). Other studies
582 have addressed the impact of pesticides on the metabolic diversity (i. e. community-level
583 physiological profiles; Foley et al. 2008, Sura et al. 2012) or the expression of certain functional genes
584 (i. e. coding carbohydrates and lignin degradation processes) of heterotrophic microbial communities
585 for the utilisation of a variety of organic carbon or nutrient sources; however these studies are few in
586 the literature (Chen et al. 2019, Su et al. 2019, Lu et al. 2020).

587 Studies show that the decomposition of leaf litter by microbial heterotrophs is sensitive to pesticides,
588 and only a few studies show neutral effects. Negative effects on litter decomposition were observed
589 when fungicides are applied, probably due to their direct effect on aquatic hyphomycete
590 communities responsible for leaf decomposition (Artigas et al. 2012b; Fernandez et al. 2015;
591 Gardestrom et al. 2016; Dawoud et al. 2017; Feckler et al. 2018b; Rossi et al. 2018). This is the case
592 for environmental concentrations of azoxystrobin and tebuconazole which were able to decrease
593 fungal biomass, litter decomposition rates and cellulolytic enzyme activities (β -glucosidase, β -

594 xylosidase ou cellobiohydrolase) (Gardestrom et al. 2016; Dawoud et al. 2017; Artigas et al. 2012b).
595 However, the extent of the effect of fungicides on microbial OM decomposition depends on: i) the
596 type of aquatic ecosystem studied (Dimitrov et al. 2014; Donnadieu et al. 2016; Pesce et al. 2016), ii)
597 the exposure history of communities to contamination (Gardestrom et al. 2016), iii) the OM quality
598 (Artigas et al. 2012b), and iv) the availability of dissolved nutrients in stream water (Rossi et al. 2018).
599 Tebuconazole can also increase certain ligninolytic enzyme activities during leaf decomposition, not
600 because of the degradation of leaf polymers, but probably because of the detoxification and/or
601 biodegradation mechanisms employed by heterotrophic components of microbial communities
602 (Rossi et al. 2018; Artigas et al. 2017). The impact of herbicides (glyphosate, Kennedy et al., 2012)
603 and insecticides (imidacloprid; Kreutzweiser et al. 2007, 2008) have been shown to weakly affect
604 microbial litter decomposition, even if slight changes in fungal biomass accumulation and/or
605 communities' composition (fungal and bacterial) are observed.

606 Very few studies have assessed the effects of pesticides in the decomposer-detritivore system, to
607 determine how pesticide effects on decomposers (bacteria and fungi) may affect detritivores
608 (macroinvertebrate shredders) based on their trophic relationships (i. e. Zubrod et al. 2011). For
609 instance, the fungicide tebuconazole has been observed to modify the structure of leaf-associated
610 microbial communities and alter the consumption of leaves by shredder macroinvertebrates (Zubrod
611 et al. 2011). Cornejo et al. (2021) showed that the decomposition of leaves in tropical streams was
612 especially reduced when fungicides and insecticides are combined (chlorpyrifos + chlorothalonil),
613 suggesting that each pesticide operates differently on microbial and macroinvertebrate communities
614 and their effects are additive. Field studies assessing pesticide impact on microbial decomposition of
615 particulate OM are also scarce, and conclusions are often contradictory. For instance, the study by
616 Brosed et al. (2016) showed that the breakdown rate of alder leaves strongly decreased along a
617 pesticide concentration gradient in 12 French streams due to effects on invertebrate's
618 decomposition but not on microbial decomposition. Piscart et al. (2011) observed that the response
619 of microbes to litter decomposition may not be sensitive enough for assessing the global effect of
620 seasonal agricultural practices. In contrast, the study of Fernandez et al. (2015) observed a strong
621 relationship between the fungicide toxicity gradient and the decrease in microbial litter
622 decomposition in 17 German streams. Rasmussen et al. (2012) observed also that microbial litter
623 decomposition was reduced by a factor of two to four in agricultural streams compared to forested
624 streams, and suggested that microbial decomposition activity responded more strongly to pesticide
625 toxicity rather than to eutrophication. Cause and effect relationships between pesticides and
626 microbial litter decomposition in the field are difficult to establish, and authors often employ "toxic
627 unit" calculations based on toxicity data (EC_{50}) from a few model organisms (i. e. microalgae) to
628 correlate with microbial decomposers activity. In view of these approaches, fungal and bacterial
629 toxicity data are needed (see Maltby et al. 2009, Ittner et al. 2018) to properly assess the impact of
630 pesticides on microbial litter decomposition in field studies.

631 The impact of pesticides has been assessed on the diversity of carbon substrates utilisation by
632 bacterial communities from rivers (Foley et al. 2008), wetlands (Sura et al. 2012; Lv et al. 2017) and
633 coastal ecosystems (Pringault et al. 2016). Two herbicide cocktails, one composed of 2,4-D, MCPA,
634 dicamba, clopyralid, dichlorprop, mecoprop, bromoxynil, glyphosate, and the other composed of
635 diuron, di-isopropyl-atrazine, 3,4-dichlorophenylurea, alachlor and linuron, altered the metabolic
636 diversity of biofilm communities from two wetlands in Manitoba, Canada (Sura et al. 2012) and
637 strongly reduced the carbohydrate utilisation (and increased that of amino acids and polymers) of
638 bacterioplankton communities from a coastal bay in Tunisia (Pringault et al. 2016). Exposure to

639 environmental concentrations of the herbicide acetochlor increased the overall quantity, but not
640 diversity, of carbon substrates utilisation by freshwater bacterial communities from US streams
641 (Foley et al. 2008). Instead, the fungicides imazalil and tebuconazole did not affect the metabolic
642 diversity of sediment bacterial communities from constructed wetlands in Denmark (Lv et al. 2017).
643 Few ecotoxicology studies have shown the impact of pesticides on metabolic diversity, which could
644 be explained by certain methodological constraints when measuring the use of carbon substrates in
645 heterotrophic microbial communities (see Preston-Mafham et al. 2002). For instance, plating may
646 induce a strong selection pressure in microbial species which would not reflect the actual
647 metabolism of microbial communities in the field. Moreover, the tetrazolium dye immediately
648 introduces some bias since not all bacteria nor fungi can reduce it, hence the plates do not
649 necessarily give a complete picture of the microbial metabolism.

650

651 *Dissipation and mitigation of contaminants and wastes*

652 The study of microbial communities' capacities for the mitigation of pesticides has been largely
653 studied in literature, mostly in the soils compared to aquatic environments (see Fenner et al. 2013).
654 However, very few studies have investigated how exposure to pesticides may affect the microbial
655 communities' capacity to biodegrade further pesticide molecules. The limited literature on this
656 subject shows that aquatic microbial communities already exposed to chronic pesticide
657 contamination in the field tend to display a higher potential to degrade certain pesticide molecules (*i.*
658 *e.* nicosulfuron; Carles et al. 2017). Nevertheless, this observation is pesticide-dependent since
659 exposure history to pesticides was overridden by phosphorus limitation in glyphosate degradation by
660 stream biofilms (Carles et al. 2019). Co-metabolism of pesticides has often been observed in
661 heterotrophic microbial communities (Liu et al. 2000). This is explained by a large number of sources
662 of naturally occurring OM available to the microbial community, a condition greatly favouring the co-
663 metabolic degradation of pesticides and the production of potentially recalcitrant metabolites
664 (Fenner et al. 2013). For instance, the biodegradation kinetics of nicosulfuron by *Plectosphaerella*
665 *cucumerina* AR1 (isolated from decomposing leaves in streams) depended on glucose concentration,
666 with a maximum specific degradation rate at 1 g/L in glucose (Carles et al. 2018). Another relevant
667 factor described by Fenner et al. (2013) is the pesticide threshold concentrations below which
668 microbial biodegradation is slowed down: this is the case of low biodegradation in groundwater
669 environments where pesticide concentrations are low and molecules can persist for longer periods.

670

671 *Dispersion of propagules*

672 The production of aquatic fungi in AME studies is mostly analysed through the counting and
673 taxonomic identification of conidia produced by aquatic hyphomycete communities (Gessner and
674 Chauvet 1994). The sporulation rates of hyphomycetes are quite sensitive to pesticides, especially
675 fungicides, with half of the studies showing a negative impact on this functional endpoint (e. g.
676 Zubrod et al. 2011; Dimitrov et al. 2014; Pimentao et al. 2020) and the other half showing neutral
677 and/or positive effects (e. g. Dawoud et al. 2017). A microcosm study showed that repeated
678 applications of the fungicide azoxystrobin decreased the sporulation rates of the hyphomycete
679 community colonizing black alder leaves, and this decrease was more marked in fungal assemblages
680 from uncontaminated forest streams compared to those from pesticide-contaminated sites
681 (Gardstrom et al. 2016). The effect of the fungicide tebuconazole on hyphomycetes sporulation

682 varied between studies, with some clear negative effects on one side (Zubrod et al. 2011; Dimitrov et
683 al. 2014; Pimentao et al. 2020) but also with some examples of stimulatory effects (Donnadieu et al.
684 2016; Dawoud et al., 2017). A possible explanation for these contradictory results may be the
685 concentration of tebuconazole used and/or the different community species composition between
686 experiments.

687 In contrast to the previous studies, Dawoud et al. (2017) observed a positive effect of high
688 tebuconazole concentrations on the sporulation of hyphomycetes which became neutral when
689 communities were contaminated by both tebuconazole and the insecticide lindane. The sporulation
690 of hyphomycetes colonizing alder leaves from a pristine stream was insensitive to the pharmaceutical
691 antifungal terbinafine (Pimentao et al. 2020). Pesticide contamination gradients in the Garonne
692 watershed (France) did not affect hyphomycete sporulation rates (Brosed et al. 2016), neither the
693 pesticide mixture composed of the fungicide imazalil and the insecticide diazinon (Flores et al. 2014).

694

695 Summarizing the main data available, pesticide exposure often impacted the following ecosystem
696 functions supported by heterotrophs in microbial communities:

697 -Carbon and nutrient regulation functions are impaired by pesticides. A reduction in litter
698 decomposition rates and a decrease in fungal biomass, is observed in microcosm studies assessing
699 fungicides toxicity. Bacterial communities are less sensitive to pesticides and often supplant fungi
700 altered by pesticides. However, these trends are less consistent in field studies where contamination
701 gradients contain not only pesticides but also nutrients, metals, pharmaceuticals, etc.

702 -Propagules dispersal (sporulation rates) in aquatic hyphomycete communities is one of the most
703 sensitive parameters to pesticide exposure in heterotrophic microbial communities, whereas
704 extracellular enzymatic activities often show transitory responses to pesticides exposure.

705 -Biotic interactions may change between decomposers and detritivores depending on the pesticide
706 applied. Despite changes in biotic interactions, the decomposition function of OM tends to remain
707 unchanged.

708

709 *3.5. Under-researched functions in AME studies*

710 A considerable number of ecosystem functions supported by microbial communities remain under-
711 researched. For instance, the gas regulation function provided by aquatic microbial communities has
712 been mostly focused on major (carbon and nitrogen) rather than on minor (i. e. sulfur) element
713 cycles. Few studies have investigated the effect of pesticides on anaerobic respiratory processes in
714 aquatic environments. For instance, OM decomposition experiments are often conducted in aerobic
715 habitats (i. e. water column or benthic surface) rather than in anaerobic habitats (i. e. hyporheic zone)
716 where OM may also accumulate. For instance, Bollinger et al. (2022) observed a twofold higher
717 fungicide effect in the hyporheic zone on microbial leaf litter decomposition compared to the benthic
718 zone. Garcia-Cruz et al. (2010) agreed on the presence of chlorophenols (i. e. 2,4-
719 dichlorophenoxyacetic acid (2,4D)) in environments where sulfate-reducing bacteria are present and
720 cause toxicity and inhibition on sulfate respiration.

721 Similar to gas cycles, most AME studies focus on pesticide effects on macro-nutrient cycles (i. e. C
722 and N cycles) rather than on micro-nutrient cycles (i. e. potassium, iron, calcium, ...). Moreover, the
723 impact of pesticides on macro-nutrient cycles is often assessed via proxies (i. e. gene expression,
724 potential enzymatic activities) rather than using tracer addition approaches (i. e. $\text{NO}_3\text{-}^{15}\text{N}$, Mulholland
725 et al. 2004) which are more integrative of microbial nutrient uptake capacities. Few studies have
726 examined the effect of pesticides on the phosphorus uptake capacity of periphyton by measuring the
727 temporal decay of soluble reactive phosphorus (SRP) (e. g. the herbicide diuron, Proia et al. 2011).
728 Further, AME studies could also integrate pesticides in carbon and nutrient cycling of contaminated
729 environments since pesticides can represent a nutrient resource for aquatic microorganisms (e. g.
730 Carles et al. 2019).

731 Overall, there is a lack of studies assessing the impacts of pesticides on the functions shared by
732 autotrophic and heterotrophic microorganisms (11% of the references explicitly consider the
733 functions as ensured by both components, see Figure 3). For instance, gas exchange (i. e. respiration),
734 OM production, nutrient regulation (i. e. nutrient uptake or enzymatic activities) or pesticide
735 dissipation functions measured at the community level reflect the sum of metabolic processes
736 carried out by both autotrophs and heterotrophs in the microbial community. Further, studies at the
737 ecosystem scale should consider the sum of metabolisms measured in the different habitats of the
738 ecosystem. Indeed, a range of studies shows the importance of considering the metabolism from
739 both planktonic and littoral habitats in lakes when assessing carbon fluxes at the ecosystem scale
740 (Vadeboncoeur et al. 2001; Vesterinen et al. 2017).

741 Finally, the protozoan community has been largely overlooked in AME studies despite contributing to
742 gas and nutrient regulation, production and supply of OM, and to the provision and maintenance of
743 biotic interactions in aquatic ecosystems. While some studies have attempted to assess pesticide
744 effect on ciliate and flagellate populations densities (e. g. Chang et al. 2011; Neury-Ormanni et al.
745 2016; Lu et al. 2020), studies assessing effects on ecosystem functions ensured by protozoans are
746 missing in the literature. Lu et al. (2020) observed that abundances of eukaryotic microbes increased
747 in the whole zooplankton community of lake Taihu (China), especially rotifers, which is in accordance
748 with the phenomenon that rotifers increase their hatchling proportion under exposure to glyphosate
749 (Gutierrez et al. 2017).

750 4. Research perspectives for AME studies based on ecosystem functions

751 This section highlights some of the research aspects missing in the literature review, that deserve
752 further investigations in order to enhance fundamental understanding in AME and improve risk
753 assessment in regard to the use of pesticides.

754 First of all, determining whether the impact of contaminants is transferred along the biological
755 continuum (from the genes to the ecosystem) will enable us to identify molecules with high
756 ecological impact. The effects of contaminants may occur at all levels of biological organization, from
757 molecular to ecosystem-level responses (Clements 2000). However, extrapolating cause-effect
758 relationships observed at the microbial population level to the community level is problematic
759 because of diverse biotic interactions (including both intra- and interspecific) and complex gradients
760 of environmental factors influencing the propagation of pesticide effects. The influence of biotic
761 interactions in leaf-associated microbial communities exposed to fungicides has been studied
762 (Artigas et al. 2017). The response of the extracellular laccase activity to the fungicide tebuconazole
763 was different when assessing the entire community response or the responses of their individual
764 components. Laccase activity of individual populations of bacteria and/or fungi was more sensitive to
765 the fungicide rather than that of the entire microbial community. The authors suggested that this
766 different response was explained by a range of biotic (i. e. species diversity and interactions) and
767 abiotic (i. e. community architecture, molecules exchange) factors avoiding the propagation of
768 pesticide effects from the population level to the community level. Similarly, the tolerance to diuron
769 was different when examining responses at the periphyton community level or at the algal
770 population level of an agricultural stream. While pollution-induced community tolerance to diuron
771 was observed for periphytic communities in the downstream site (more contaminated) compared to
772 those from the upstream site (less contaminated), certain diatom strains (i. e. *Encyonema*
773 *neomesianum*) collected from the downstream site were more sensitive than those collected from
774 the upstream site (Roubeix et al. 2012). The authors from this study suggested that these differences
775 were partly explained by the co-occurrence *in situ* of copper and diuron contamination gradients, *E.*
776 *neomesianum* strains from downstream being more tolerant to copper. Extrapolation of pesticide
777 effects between levels of biological organisation is thus tricky given the environmental context of
778 multi-contamination. Beyond these studies, identifying "keystone species" within microbial
779 communities and testing the effect of pesticides on these specific populations could be a research
780 strategy to better address the propagation of pesticide effects at the community level or even the
781 entire ecosystem.

782 Second, we observed a very low number of references (7%) considering explicitly pesticide effects on
783 the autotrophic and heterotrophic components at the same time, permitting to have a more
784 integrated view of the overall response of the aquatic microbial community to contaminants.
785 Assessing simultaneously functions ensured by both autotrophs and heterotrophs could be a way to
786 better integrate the impacts of pesticides on the entire ecosystem functioning. Such a research
787 challenge could be overcome by the miniaturization of tests permitting to process a large number of
788 samples. Tests in microplates are already used for the assessment of pesticide effects on microbial
789 photosynthesis (e. g. Gardia-Parège, Kim-Tiam et al. 2022), or carbon substrate use (e. g. Foley et al.
790 2008, Sura et al. 2012, Pringault et al. 2016, Lv et al. 2017). Moreover, studies permitting to have a
791 more integrated response of the ecosystem (i. e. trophic chains such as decomposers-detritivores-
792 carnivores or primary producers-herbivores-carnivores) are rare in the literature. Some studies on
793 leaf litter decomposition observed that the contamination by pesticides does not impair microbial-
794 mediated decomposition but often decreases invertebrate-mediated decomposition (Piscart et al.

795 2011; Brosted et al. 2016; Rossi et al. 2019; Jabiol et al. 2022). Despite such a top-down effect of
796 pesticides in the decomposer-detrivore system, other studies have shown bottom-up effects in the
797 producer-consumer system explained by pesticides' effect on food resources quality (rarefaction of
798 species rich in omega-3 and 6 fatty acids) to consumers (see sections "Provision and maintenance of
799 biotic interactions" in 3.3 and 3.4). Microbial communities can bioaccumulate pesticides (see sections
800 "Dissipation and mitigation of contaminants" in 3.3 and 3.4) and contribute to their transfer through
801 the trophic chain. In turn, consumers can also adapt their diet as a function of the content (nutritious
802 quality and amount of contaminants) by avoiding some unsuitable food sources or diversifying them
803 (Neury-Ormanni et al. 2020, Bundschuh et al. 2011). This topic certainly deserves more research by
804 comparing different types of exposure conditions, cocktails of molecules (including metabolites), and
805 trophic interactions.

806 Third, examining the species selection and their physiological and metabolic adaptations in
807 environments chronically contaminated by pesticides will permit us to determine the recovery
808 potential of these communities to a non-contaminated status. Both autotrophic and heterotrophic
809 components of aquatic microbial communities have shown strong functional adaptability of
810 microbial communities chronically exposed to pesticides, even if this contradicts the literature survey
811 by Allison and Martigny (2008) in which microbial communities are sensitive to disturbance and often
812 do not rapidly recover to their original state after chronic exposure. Blanck (2002) described this
813 adaptation process based on profound structural changes in microbial communities' composition, i. e.
814 the elimination of sensitive species followed by the development of pesticide-tolerant species in the
815 microbial community, in his so-called "pollution-induced community tolerance" concept. However,
816 structural changes in pesticide-polluted communities are not always accompanied by functional
817 changes and this is because tolerant species can compensate for the loss of sensitive competitors
818 and maintain ecosystem functions. Unfortunately, the knowledge of the presence or absence of
819 sensitive and tolerant species offers little information on their functional capacities, since strong
820 functional redundancy exists in microbial communities (see Tlili et al. 2016). The study of Feckler et al.
821 (2018a) showed comparatively minor adverse effects of fungicides or even stimulation of ecosystem
822 functions in microbial communities previously exposed to agricultural contamination. Similar results
823 were observed by Pesce et al. (2010) with the effect of the herbicide diuron in periphyton
824 communities chronically exposed to pesticide contamination. Research efforts are still needed to
825 better establish the functional traits characteristic of microbial species (Bier et al. 2015) and their
826 responses to pesticides. Beyond that, the study of genetic mutations in microorganisms resulting
827 from selection pressure due to exposure to pesticides is poorly investigated in aquatic microbial
828 communities, compared to soils (e. g. Pileggi et al. 2020). Understanding the versatility of microbial
829 species to adapt to pesticides and their functional role will certainly help to understand the
830 functioning of microbial communities in environments chronically contaminated by pesticides.

831 Fourth, unravelling "omics" multi-functional responses of microbial communities to pesticides will
832 allow us to detect, through non-targeted analyses, unsuspected functional responses to pesticides.
833 The literature review of Ebner (2021) found only 648 studies addressing "omics" and "ecotoxicology"
834 in the last twenty years (2000-2020) which is a relatively low score. This review also concludes that
835 transcriptomics is the most frequently applied method (43%), followed by proteomics (30%),
836 metabolomics (13%) and finally, multi-omics (combination of two or multiple omics methods, 13%).
837 We still identify a gap in the potential use of omics data to explain multiple functions in microbial
838 communities. Johnson et al. (2015) pioneered in suggesting an "association mining approach"
839 between the rate constants of observed biotransformation reactions and meta-omics data as an

840 untargeted approach to generate hypotheses about potential causal linkages between enzymes and
841 pesticides biotransformation. Recently, Achermann et al. (2020) used association mining to
842 demonstrate quantitative correlations between metatranscriptomic data and micropollutant
843 biotransformation in activated sludge. We consider that extrapolation of association mining between
844 chemical and metatranscriptomic profiling to enzymes likely involved in catalysing reactions related
845 to important ecosystem functions, under natural conditions, would help in bridging the gap between
846 -omics data potential and the wide diversity of functions ensured by microbial communities.

847

848 **5. Concluding remarks**

849 As highlighted by this literature review of the last 20 years of AME research, ecosystem functions
850 ensured by aquatic microbial communities are threatened by pesticide contamination. It is important
851 to remark that the assessment of pesticide effects on microbial functions is based on a limited
852 number of pesticide molecules (mainly, herbicides for autotrophs and fungicides for heterotrophs)
853 which does not allow for a complete picture of the multiple contaminations affecting aquatic
854 microbial communities. Surprisingly, AME works that focused on autotrophs mostly studied aspects
855 related to the functions of provision and maintenance of biodiversity and biotic interactions, while
856 AME studies on heterotrophs mostly examine functions related to carbon and nutrient regulation.
857 Integrative ecotoxicological risk assessment from microbial communities to ecosystem functions calls
858 for more complex experiments and requires exchanges between more or less distant fields of
859 research. We believe that incorporating measurements of function, as often done in ecology
860 approaches, would put forward ecosystemic consequences of pesticide exposure and allow to some
861 extent the quantification of the impacts of pesticide pollution on ecosystem functioning. Obtaining
862 such functional data at large spatial and temporal scales is necessary to raise public awareness and
863 likely capture the attention of the operational sphere, and thus promote the use of aquatic microbial
864 community functioning in regulatory frameworks (including Ecological Risk Assessment).

865

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867

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879

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881

882 **7. References**

883 Abelho, M., Martins, T.F., Shinn, C., Moreira-Santos, M., Ribeiro, R. (2016). Effects of the fungicide
884 pyrimethanil on biofilm and organic matter processing in outdoor lentic mesocosms.
885 *Ecotoxicology* 25, 121–131.

886 Achermann, S., Mansfeldt, C. B., Müller, M., Johnson, D. R., Fenner, K. (2020). Relating
887 metatranscriptomic profiles to the micropollutant biotransformation potential of complex
888 microbial communities. *Environmental Science & Technology*, 54, 235-244.

889 Allison, S. D., Martiny, J. B. (2008). Resistance, resilience, and redundancy in microbial communities.
890 *Proceedings of the National Academy of Sciences* 105, 11512-11519.

891 Artigas, J., Batisson, I., Carles, L. (2020). Dissolved organic matter does not promote glyphosate
892 degradation in auto-heterotrophic aquatic microbial communities. *Environmental Pollution*
893 259, 113951.

894 Artigas, J., Rossi, F., Gerphagnon, M., Mallet, C. (2017). Sensitivity of laccase activity to the fungicide
895 tebuconazole in decomposing litter. *Science of the Total Environment*, 584, 1084-1092.

896 Artigas, J., Pascault, N., Bouchez, A., Chastain, J., Debroas, D., Humbert, J.F., Leloup, J., Tadonleke,
897 R.D., ter Halle, A., Pesce, S. (2014). Comparative sensitivity to the fungicide tebuconazole of
898 biofilm and plankton microbial communities in freshwater ecosystems. *Science of the Total*
899 *Environment* 468, 326-336.

900 Artigas, J., Arts, G., Babut, M., Caracciolo, A., Charles, S. ... (2012a). Towards a renewed research
901 agenda in ecotoxicology. *Environmental Pollution* 160, 201-207.

902 Artigas, J., Majerholc, J., Foulquier, A., Margoum, C., Volat, B., Neyra, M., Pesce, S. (2012b). Effects of
903 the fungicide tebuconazole on microbial capacities for litter breakdown in streams. *Aquatic*
904 *Toxicology* 122, 197-205.

905 Battin, T. J., Kaplan, L. A., Denis Newbold, J., & Hansen, C. M. (2003). Contributions of microbial
906 biofilms to ecosystem processes in stream mesocosms. *Nature*, 426, 439-442.

907 Bayona, Y., Roucaute, M., Cailleaud, K., Lagadic, L., Basseres, A., Caquet, T. (2014). Structural and
908 biological trait responses of diatom assemblages to organic chemicals in outdoor flow-
909 through mesocosms. *Environmental Pollution* 192, 186-195.

910 Bérard, A., Dorigo, U., Mercier, I., Becker-van Slooten, K., Grandjean, D., Leboulanger, C. (2003).
911 Comparison of the ecotoxicological impact of the triazines Irgarol 1051 and atrazine on
912 microalgal cultures and natural microalgal communities in Lake Geneva. *Chemosphere* 53,
913 935-944.

914 Bier, R. L., Bernhardt, E. S., Boot, C. M., Graham, E. B., Hall, E. K., Lennon, J. T., et al. (2015). Linking
915 microbial community structure and microbial processes: An empirical and conceptual
916 overview. *FEMS Microbiology Ecology* 91, fiv113.

917 Bighiu, M.A., Hoss, S., Traunspurger, W., Kahlert, M., Goedkoop, W. (2020). Limited effects of
918 pesticides on stream macroinvertebrates, biofilm nematodes, and algae in intensive
919 agricultural landscapes in Sweden. *Water Research* 174, 115640.

920 Blanck, H. (2002). A critical review of procedures and approaches used for assessing pollution-
921 induced community tolerance (PICT) in biotic communities. *Human and Ecological Risk*
922 *Assessment*, 8, 1003–1034.

923 Bonnineau, C., Artigas, J., Chaumet, B., Dabrin, A., Faburé, J., Ferrari, B.J.D., Lebrun, J.D., Margoum,
924 C., Mazzella, N., Miège, C., Morin, S., Uher, E., Babut, M., Pesce, S. (2021). Role of Biofilms in
925 Contaminant Bioaccumulation and Trophic Transfer in Aquatic Ecosystems: Current State of
926 Knowledge and Future Challenges. *Reviews of Environmental Contamination and Toxicology*
927 253, 115-153.

928 Bollinger, E., Zubrod, J. P., Kanschak, M., Sulzer, L., Schnurr, J., Schreiner, V. C., ... Bundschuh, M.
929 (2022). As above, so below? Effects of fungicides on microbial organic matter decomposition
930 are stronger in the hyporheic than in the benthic zone. *Limnology and Oceanography* 67, 39-
931 52.

932 Brêda-Alves, F., de Oliveira Fernandes, V., Chia, M.A. (2021). Understanding the environmental roles
933 of herbicides on cyanobacteria, cyanotoxins, and cyanoHABs. *Aquatic Ecology* 55, 347–361.

934 Briand, E., Pringault, O., Jacquet, S., & Torrétion, J. P. (2004). The use of oxygen microprobes to
935 measure bacterial respiration for determining bacterioplankton growth efficiency. *Limnology*
936 *and Oceanography: Methods* 2, 406-416.

937 Bricheux, G., Le Moal, G., Hennequin, C., Coffe, G., Donnadiou, F., Portelli, C., Bohatier, J., Forestier,
938 C. (2013). Characterization and evolution of natural aquatic biofilm communities exposed in
939 vitro to herbicides. *Ecotoxicology and Environmental Safety* 88, 126-134.

940 Brosed, M., Lamothe, S., Chauvet, E. (2016). Litter breakdown for ecosystem integrity assessment
941 also applies to streams affected by pesticides. *Hydrobiologia* 773, 87-102

942 Bundschuh, M., Zubrod, J. P., Kosol, S., Maltby, L., Stang, C., Duester, L., Schulz, R. (2011). Fungal
943 composition on leaves explains pollutant-mediated indirect effects on amphipod feeding.
944 *Aquatic Toxicology* 104, 32-37.

945 Campbell, C. D., Chapman, S. J., Cameron, C. M., Davidson, M. S., & Potts, J. M. (2003). A rapid
946 microtiter plate method to measure carbon dioxide evolved from carbon substrate
947 amendments so as to determine the physiological profiles of soil microbial communities by
948 using whole soil. *Applied and Environmental Microbiology*, 69, 3593-3599.

949 Carles, L., Artigas, J. (2020). Interaction between glyphosate and dissolved phosphorus on bacterial
950 and eukaryotic communities from river biofilms. *Science of the Total Environment* 719,
951 137463.

952 Carles, L., Gardon, H., Joseph, L., Sanchís, J., Farre, M., Artigas, J. (2019). Meta-analysis of glyphosate
953 contamination in surface waters and dissipation by biofilms. *Environment international* 124,
954 284-293.

955 Carles, L., Rossi, F., Besse-Hoggan, P., Blavignac, C., Lereboure, M., Artigas, J., Batisson, I. (2018).
956 Nicosulfuron degradation by an ascomycete fungus isolated from submerged *Alnus* leaf litter.
957 *Frontiers in Microbiology* 9, 3167.

958 Carles, L., Rossi, F., Joly, M., Besse-Hoggan, P., Batisson, I., Artigas, J. (2017). Biotransformation of
959 herbicides by aquatic microbial communities associated to submerged leaves. *Environmental*
960 *Science and Pollution Research* 24, 3664-3674.

961 Chang, K.H., Sakamoto, M., Ha, J.Y., Miyabara, Y., Nakano, S., Doi, H., Hanazato, T. (2011). Response
962 of the plankton community to herbicide application (triazine herbicide, simetryn) in a
963 eutrophicated system: short-term exposure experiment using microcosms. *Limnology* 12, 11–
964 16.

965 Chaumet, B., Morin, S., Hourtané, O., Artigas, J., Delest, B., Eon, M., Mazzella, N. (2019). Flow
966 conditions influence diuron toxicokinetics and toxicodynamics in freshwater biofilms. *Science*
967 *of The Total Environment* 652, 1242-1251.

968 Chen, Y., Su, X.X., Wang, Y.Y., Zhao, S.Y., He, Q. (2019). Short-term responses of denitrification to
969 chlorothalonil in riparian sediments: Process, mechanism and implication. *Chemical*
970 *Engineering Journal* 358, 1390-1398.

971 Clements, W. H. (2000) Integrating effects of contaminants across levels of biological organization: an
972 overview. *Journal of Aquatic Ecosystem Stress and Recovery* 7, 113–116.

973 Cornejo, A., Pérez, J., López-Rojo, N., García, G., Pérez, E., Guerra, A., ... Boyero, L. (2021). Litter
974 decomposition can be reduced by pesticide effects on detritivores and decomposers:
975 Implications for tropical stream functioning. *Environmental Pollution* 285, 117243.

976 Coste, M., Boutry, S., Tison-Rosebery, J., Delmas, F. (2009). Improvements of the Biological Index
977 (BDI): description and efficiency of the new version (BDI-2006). *Ecological Indicators* 9,621-
978 650.

979 Cothran, R.D., Radarian, F., Relyea, R.A. (2011). Altering aquatic food webs with a global insecticide:
980 arthropod–amphibian links in mesocosms that simulate pond communities. *Journal of the*
981 *North American Benthological Society* 30, 893-912.

982 Cycoń, M., Piotrowska-Seget, Z., Kaczyńska, A., Kozdrój, J. (2006). Microbiological characteristics of a
983 sandy loam soil exposed to tebuconazole and λ -cyhalothrin under laboratory conditions.
984 *Ecotoxicology* 15, 639-646.

985 Dawoud, M., Bundschuh, M., Goedkoop, W., McKie, B.G. (2017). Interactive effects of an insecticide
986 and a fungicide on different organism groups and ecosystem functioning in a stream detrital
987 food web. *Aquatic Toxicology* 186, 215-221.

988 de la Broise, D., Stachowski-Haberkorn, S. (2012). Evaluation of the partial renewal of in situ
989 phytoplankton microcosms and application to the impact assessment of bentazon and
990 dimethenamid. *Marine Pollution Bulletin* 64, 2480-2488.

991 De Groot, R. S., Wilson, M. A., & Boumans, R. M. (2002). A typology for the classification, description
992 and valuation of ecosystem functions, goods and services. *Ecological economics*, 41, 393-408.

993 De Stefano, L.G., Gattas, F., Vinocur, A., Cristos, D., Rojas, D., Cataldo, D., Pizarro, H. (2018).
994 Comparative impact of two glyphosate-based formulations in interaction with *Limnoperna*
995 *fortunei* on freshwater phytoplankton. *Ecological Indicators* 85, 575-584.

996 Debenest, T., Silvestre, J., Coste, M., Pinelli, E. (2010). Effects of pesticides on freshwater diatoms.
997 *Reviews of Environmental Contamination and Toxicology* 203, 87-103.

998 Debenest, T., Pinelli, E., Coste, M., Silvestre, J., Mazzella, N., Madigou, C., Delmas, F. (2009).
999 Sensitivity of freshwater periphytic diatoms to agricultural herbicides. *Aquatic Toxicology* 93,
1000 11-17.

1001 Demailly, F., Elfeky, I., Malbezin, L., Le Guédard, M., Eon, M., Bessoule, J.J., Feurtet-Mazel, A.,
1002 Delmas, F., Mazzella, N., Gonzalez, P., Morin, S. (2019). Impact of diuron and S-metolachlor
1003 on the freshwater diatom *Gomphonema gracile*: Complementarity between fatty acid
1004 profiles and different kinds of ecotoxicological impact-endpoints. *Science of the Total*
1005 *Environment* 688, 960-969.

1006 Dimitrov, M.R., Kosol, S., Smidt, H., Buijse, L., Van den Brink, P.J., Van Wijngaarden, R.P.A., Brock,
1007 T.C.M., Maltby, L. (2014). Assessing effects of the fungicide tebuconazole to heterotrophic
1008 microbes in aquatic microcosms. *Science of the Total Environment* 490, 1002- 1011.

1009 Donnadieu, F., Besse-Hoggan, P., Forestier, C., Artigas, J. (2016). Influence of streambed substratum
1010 composition on stream microbial communities exposed to the fungicide tebuconazole.
1011 *Freshwater Biology* 61, 2026-2036.

1012 Dorigo, U., Berard, A., Rimet, F., Bouchez, A., Montuelle, B. (2010). In situ assessment of periphyton recovery in a river contaminated by pesticides.
1013 *Aquatic Toxicology* 98, 396-406.

1014 Dorigo, U., Berard, A., Rimet, F., Bouchez, A., Montuelle, B. (2010). In situ assessment of periphyton
1015 recovery in a river contaminated by pesticides. *Aquatic Toxicology* 98, 396-406.

1016 Dorigo, U., Lefranc, M., Leboulanger, C., Montuelle, B., Humbert, J.F. (2009). Spatial heterogeneity of
1017 periphytic microbial communities in a small pesticide-polluted river. *FEMS Microbiology*
1018 *Ecology* 67, 491-501.

1019 Dorigo, U., Leboulanger, C., Berard, A., Bouchez, A., Humbert, J.F., Montuelle, B. (2007). Lotic biofilm
1020 community structure and pesticide tolerance along a contamination gradient in a vineyard
1021 area. *Aquatic Microbial Ecology* 50, 91–102.

1022 Ebner, J. N. (2021). Trends in the application of “omics” to ecotoxicology and stress
1023 ecology. *Genes*, 12, 1481.

1024 Feckler, A., Bundschuh, M. (2020). Decoupled structure and function of leaf-associated
1025 microorganisms under anthropogenic pressure: Potential hurdles for environmental
1026 monitoring. *Freshwater Science*, 39, 652-664.

1027 Feckler, A., Goedkoop, W., Kenschak, M., Bundschuh, R., Kenngott, K.G.J., Schulz, R., Zubrod, J.P.,
1028 Bundschuh, M. (2018a). History matters: Heterotrophic microbial community structure and
1029 function adapt to multiple stressors. *Global Change Biology* 24, E402-E415.

1030 Feckler, A., Rakovic, J., Kahlert, M., Troger, R., Bundschuh, M. (2018b). Blinded by the light: Increased
1031 chlorophyll fluorescence of herbicide-exposed periphyton masks unfavorable structural
1032 responses during exposure and recovery. *Aquatic Toxicology* 203, 187-193.

1033 Feckler, A., Goedkoop, W., Zubrod, J. P., Schulz, R., Bundschuh, M. (2016). Exposure pathway-
1034 dependent effects of the fungicide epoxiconazole on a decomposer-detritivore system.
1035 *Science of the Total Environment* 571, 992-1000.

1036 Fenner, K., Canonica, S., Wackett, L. P., & Elsner, M. (2013). Evaluating pesticide degradation in the
1037 environment: blind spots and emerging opportunities. *Science* 341, 752-758.

1038 Fernandez, D., Voss, K., Bundschuh, M., Zubrod, J.P., Schafer, R.B. (2015). Effects of fungicides on
1039 decomposer communities and litter decomposition in vineyard streams. *Science of the Total*
1040 *Environment* 533, 40-48.

1041 Field, C.B., Behrenfeld, M.J., Randerson, J.T., Falkowski, P. (1998). Primary production of the
1042 biosphere: integrating terrestrial and oceanic components. *Science* 281, 237-240.

1043 Filimonova, V., Nys, C., De Schamphelaere, K.A.C., Goncalves, F., Marques, J.C., Goncalves, A.M.M.,
1044 De Troch, M. (2018). Ecotoxicological and biochemical mixture effects of an herbicide and a
1045 metal at the marine primary producer diatom *Thalassiosira weissflogii* and the primary
1046 consumer copepod *Acartia tonsa*. *Environmental Science and Pollution Research* 25, 22180–
1047 22195.

1048 Filimonova, V., Goncalves, F., Marques, J.C., De Troch, M., Goncalves, A.M.M. (2016). Biochemical
1049 and toxicological effects of organic (herbicide Primextra Gold TZ) and inorganic (copper)
1050 compounds on zooplankton and phytoplankton species. *Aquatic Toxicology* 177, 33-43.

1051 Flores, L., Banjac, Z., Farre, M., Larranaga, A., Mas-Marti, E., Munoz, I., Barcelo, D., Elosegí, A. (2014).
1052 Effects of a fungicide (imazalil) and an insecticide (diazinon) on stream fungi and
1053 invertebrates associated with litter breakdown. *Science of the Total Environment* 476, 532-
1054 541.

1055 Friberg-Jensen, U., Wendt-Rasch, L., Woin, P., Christoffersen, K. (2003). Effects of the pyrethroid
1056 insecticide, cypermethrin, on a freshwater community studied under field conditions. I.
1057 Direct and indirect effects on abundance measures of organisms at different trophic levels.
1058 *Aquatic Toxicology* 63, 357-371.

1059 Foley, M.E., Sigler, V., Gruden, C.L. (2008). A multiphasic characterization of the impact of the
1060 herbicide acetochlor on freshwater bacterial communities. *ISME Journal* 2, 56-66.

1061 Garcia-Cruz, U., Celis, L.B., Poggi, H., Meraz, M. (2010). Inhibitory concentrations of 2,4D and its
1062 possible intermediates in sulfate reducing biofilms. *Journal of Hazardous Materials* 179, 591-
1063 595.

1064 Garcia-Ortega, S., Holliman, P.J., Jones, D.L. (2011). Effects of salinity, DOM and metals on the fate
1065 and microbial toxicology of propetamphos formulations in river and estuarine sediment.
1066 *Chemosphere* 83, 1117-1123.

1067 Gardstrom, J., Ermold, M., Goedkoop, W., McKie, B.G. (2016). Disturbance history influences
1068 stressor impacts: effects of a fungicide and nutrients on microbial diversity and litter
1069 decomposition. *Freshwater Biology* 61, 2171-2184.

1070 Gardia-Parège, C., Kim-Tiam, S., Budzinski, H., Mazzella, N., Devier, M.H., Morin, S. (2022). Pesticide
1071 toxicity towards microalgae increases with environmental mixture complexity. *Environmental*
1072 *Science and Pollution Research* 29, 29368-29381.

1073 Gattás, F., De Stefano, L.G., Vinocur, A., Bordet, F., Espinosa, M.S., Pizarro, H., Cataldo, D. (2018).
1074 Impact of interaction between *Limnoperna fortunei* and Roundup Max (R) on freshwater
1075 phytoplankton: An in situ approach in Salto Grande reservoir (Argentina). *Chemosphere* 209,
1076 748-757.

1077 Gessner, M. O., Chauvet, E. (1994). Importance of stream microfungi in controlling breakdown rates
1078 of leaf litter. *Ecology*, 75, 1807-1817.

1079 Ghiglione, J., Martin-Laurent, F., Pesce, S. (2016). Microbial ecotoxicology: an emerging discipline
1080 facing contemporary environmental threats. *Environmental Science and Pollution Research*
1081 23, 3981–3983.

1082 Gonçalves, A.M.M., Mesquita, A.F., Verdelhos, T., Coutinho, J.A.P., Marques, J.C., Gonçalves, F.
1083 (2016). Fatty acids' profiles as indicators of stress induced by of a common herbicide on two
1084 marine bivalves species: *Cerastoderma edule* (Linnaeus, 1758) and *Scrobicularia plana* (da
1085 Costa, 1778). *Ecological Indicators* 63, 209-218.

1086 Gustavson, K., Møhlenberg, F., Schlüter, L. (2003). Effects of Exposure Duration of Herbicides on
1087 Natural Stream Periphyton Communities and Recovery. *Archives of Environmental*
1088 *Contamination and Toxicology* 45, 48–58.

1089 Gutierrez, M. F., Battauz, Y., & Caisso, B. (2017). Disruption of the hatching dynamics of zooplankton
1090 egg banks due to glyphosate application. *Chemosphere*, 171, 644-653.

1091 Hayashi, S., Itoh, K., Suyama, K. (2011). Evaluation of river model biofilm for assessing pesticide
1092 effects: a case study with atrazine. *Journal of Pesticide Science* 36, 243-247.

1093 Hua, J., Relyea, R.A. (2012). East Coast vs West Coast: effects of an insecticide in communities
1094 containing different amphibian assemblages. *Freshwater Science* 31, 787-799.

1095 Ittner, L. D., Junghans, M., & Werner, I. (2018). Aquatic fungi: a disregarded trophic level in ecological
1096 risk assessment of organic fungicides. *Frontiers in Environmental Science*, 6, 105.lummato,
1097 M.M., Pizarro, H., Cataldo, D., Di Fiori, E., Afonso, M.D., de Molina, M.D.R., Juarez, A.B.
1098 (2017). Effect of glyphosate acid on biochemical markers of periphyton exposed in outdoor
1099 mesocosms in the presence and absence of the mussel *Limnoperna fortunei*. *Environmental*
1100 *Toxicology and Chemistry* 36, 1775-1784.

1101 Jabiol, J., Chauvet, E., Guérol, F., Bouquerel, J., Usseglio-Polatera, P., Artigas, J., ... Gouy, V. (2022).
1102 The combination of chemical, structural, and functional indicators to evaluate the
1103 anthropogenic impacts on agricultural stream ecosystems. *Environmental Science and*
1104 *Pollution Research* 29, 29296-29313.

1105 Johnson, D. R., Helbling, D. E., Men, Y., Fenner, K. (2015). Can meta-omics help to establish causality
1106 between contaminant biotransformations and genes or gene products? *Environ. Sci.: Water*
1107 *Res. Technol.* 1, 272– 278.

1108 Kennedy, E., Leff, L.G., de Szalay, F.A. (2012). Herbiciding *Phragmites australis*: effects on litter
1109 decomposition, microbial biomass, and macroinvertebrate communities. *Fundamental and*
1110 *Applied Limnology* 180, 309-319.

1111 Khadra, M., Planas, D., Girard, C., Amyot, M. (2018). Age matters: Submersion period shapes
1112 community composition of lake biofilms under glyphosate stress. *FACETS* 3, 934-951.

1113 Kim-Tiam, S., Morin, S., Bonet, B., Guasch, H., Feurtet-Mazel, A., Eon, M., Gonzalez, P., Mazzella, N.
1114 (2015). Is the toxicity of pesticide mixtures on river biofilm accounted for solely by the major
1115 compounds identified? *Environmental Science and Pollution Research* 22, 4009-4024.

1116 Kim-Tiam, S., Morin, S., Pesce, S., Feurtet-Mazel, A., Moreira, A., Gonzalez, P., Mazzella, N. (2014).
1117 Environmental effects of realistic pesticide mixtures on natural biofilm communities with
1118 different exposure histories. *Science of The Total Environment* 473-474, 496-506.

1119 Knapp, C. W., Caquet, T., Hanson, M. L., Lagadic, L., Graham, D. W. (2005). Response of water column
1120 microbial communities to sudden exposure to deltamethrin in aquatic mesocosms. *FEMS*
1121 *Microbiology Ecology* 54, 157-165.

1122 Kreuzweiser, D.P., Good, K.P., Chartrand, D.T., Scarr, T.A., Thompson, D.G. (2008). Toxicity of the
1123 systemic insecticide, imidacloprid, to forest stream insects and microbial communities.
1124 *Bulletin of Environmental Contamination and Toxicology* 80, 211-214.

1125 Kreuzweiser, D., Good, K., Chartrand, D., Scarr, T., Thompson, D. (2007). Non-target effects on
1126 aquatic decomposer organisms of imidacloprid as a systemic insecticide to control emerald
1127 ash borer in riparian trees. *Ecotoxicology and Environmental Safety* 68, 315-325.

1128 Kumar, A., Singh, J.S. (2017). Cyanoremediation: A Green-Clean Tool for Decontamination of
1129 Synthetic Pesticides from Agro- and Aquatic Ecosystems. In: Singh, J., Seneviratne, G. (eds)
1130 *Agro-Environmental Sustainability*. Springer, Cham.

1131 Larras, F., Coulaud, R., Gautreau, E., Billoir, E., Rosebery, J., Usseglio-Polatera, P. (2017). Assessing
1132 anthropogenic pressures on streams: A random forest approach based on benthic diatom
1133 communities. *Science of The Total Environment* 586, 1101-1112.

1134 Larras, F., Gregorio, V., Bouchez, A., Montuelle, B., Chevre, N. (2016). Comparison of specific versus
1135 literature species sensitivity distributions for herbicides risk assessment. *Environmental*
1136 *Science and Pollution Research* 23, 3042-3052.

1137 Lawrence, J.R., Kopf, G., Headley, J.V., Neu, T.R. (2001). Sorption and metabolism of selected
1138 herbicides in river biofilm communities. *Canadian Journal of Microbiology* 47, 634-641.

1139 Leenhardt, S., Mamy, L., Pesce, S., Sanchez, W. (2022). The impacts of plant protection products on
1140 biodiversity and ecosystem services. Summary of the collective scientific expertise. INRAE-
1141 IFREMER, 14 p. { DOI : 10.17180/d7a0-f411}

1142 Liu, D., Maguire, R. J., Lau, Y. L., Pacepavicius, G. J., Okamura, H., Aoyama, I. (2000). Factors affecting
1143 chemical biodegradation. *Environmental Toxicology* 15, 476-483.

1144 Lock, M. A., Wallace, R. R., Costerton, J. W., Ventullo, R. M., & Charlton, S. E. (1984). River epilithon:
1145 toward a structural-functional model. *Oikos*, 10-22.

1146 López-Doval, J.C., Ricart, M., Guasch, H., Román, A.M., Sabater, S., Muñoz, I. (2010). Does Grazing
1147 Pressure Modify Diuron Toxicity in a Biofilm Community? *Archives of Environmental*
1148 *Contamination and Toxicology* 58, 955-962.

1149 Lozano, V.L., Vinocur, A., Garcia, C.A.S.Y., Allende, L., Cristos, D.S., Rojas, D., Wolansky, M., Pizarro, H.
1150 (2018). Effects of glyphosate and 2,4-D mixture on freshwater phytoplankton and periphyton
1151 communities: a microcosms approach. *Ecotoxicology and Environmental Safety* 148, 1010-
1152 1019.

1153 Lu, T., Xu, N.H., Zhang, Q., Zhang, Z.Y., Debognies, A., Zhou, Z.G., Sun, L.W., Qian, H.F. (2020).
1154 Understanding the influence of glyphosate on the structure and function of freshwater
1155 microbial community in a microcosm. *Environmental Pollution* 260, 9.

1156 Lu, T., Zhang, Q., Lavoie, M., Zhu, Y., Ye, Y., Yang, J., Paerl, H.W., Qian, H., Zhu, Y.G. (2019). The
1157 fungicide azoxystrobin promotes freshwater cyanobacterial dominance through altering
1158 competition. *Microbiome* 7, 128.

1159 Lüring, M., Roessink, I. (2006). On the way to cyanobacterial blooms: Impact of the herbicide
1160 metribuzin on the competition between a green alga (*Scenedesmus*) and a cyanobacterium
1161 (*Microcystis*). *Chemosphere* 65, 618-626.

1162 Lv, T., Zhang, Y., Carvalho, P.N., Zhang, L., Button, M., Arias, C.A., Weber, K.P., Brix, H. (2017).
1163 Microbial community metabolic function in constructed wetland mesocosms treating the
1164 pesticides imazalil and tebuconazole. *Ecological Engineering* 98, 378-387.

1165 Magnusson, M., Heimann, K., Ridd, M., Negri, A.P. (2012). Chronic herbicide exposures affect the
1166 sensitivity and community structure of tropical benthic microalgae. *Marine Pollution Bulletin*
1167 65, 363-372.

1168 Magnusson, M., Heimann, K., Ridd, M., Negri, A.P. (2013). Pesticide contamination and phytotoxicity
1169 of sediment interstitial water to tropical benthic microalgae. *Water Research* 47, 5211-5221.

1170 Maltby, L., Brock, T. C., Van den Brink, P. J. (2009). Fungicide risk assessment for aquatic ecosystems:
1171 importance of interspecific variation, toxic mode of action, and exposure regime.
1172 *Environmental Science & Technology* 43, 7556-7563.

1173 Marcel, R., Bouchez, A., Rimet, F. (2013). Influence of herbicide contamination on diversity and
1174 ecological guilds of river diatoms. *Cryptogamie Algologie* 34, 169-183.

1175 Matz, C., Kjelleberg, S. (2005). Off the hook—how bacteria survive protozoan grazing. *Trends in*
1176 *Microbiology* 13, 302-307.

1177 Mbanaso, F.U., Coupe, S.J., Charlesworth, S.M., Nnadi, E.O., Ifelebuegu, A.O. (2014). Potential
1178 microbial toxicity and non-target impact of different concentrations of glyphosate-containing
1179 herbicide (GCH) in a model pervious paving system. *Chemosphere* 100, 34-41.

1180 Milenkovski, S., Baath, E., Lindgren, P.E., Berglund, O. (2010). Toxicity of fungicides to natural
1181 bacterial communities in wetland water and sediment measured using leucine incorporation
1182 and potential denitrification. *Ecotoxicology* 19, 285-294.

1183 Moreira-Santos, M., da Silva, E.M., Soares, A.M.V.M., Ribeiro, R. (2005). In situ and laboratory
1184 microalgal assays in the tropics: A microcosm simulation of edge-of-field pesticide runoff.
1185 *Bulletin of Environmental Contamination and Toxicology* 74, 48–55.

1186 Morin, S., Gómez, N., Tornes, E., Licursi, M., Tison-Rosebery, J. (2016). Benthic diatom monitoring
1187 and assessment of freshwater environments: Standard methods and future challenges. In:
1188 A.M. Romani; H. Guasch; M.D. Balaguer. *Aquatic Biofilms: Ecology, Water Quality and*
1189 *Wastewater Treatment*, Caister Academic Press, 111-124.

1190 Morin, S., Pesce, S., Tlili, A., Coste, M., Montuelle, B. (2010). Recovery potential of periphytic
1191 communities in a river impacted by a vineyard watershed. *Ecological Indicators* 10, 419-426.

1192 Morin, S., Bottin, M., Mazzella, N., Macary, F., Delmas, F., Winterton, P., Coste, M. (2009). Linking
1193 diatom community structure to pesticide input as evaluated through a spatial contamination
1194 potential (Phytopixal): a case study in the Neste river system (South-West France). *Aquatic*
1195 *Toxicology* 94, 28-39.

1196 Mulholland P.J., Valett H.M., Webster J.R., Thomas S.A., Cooper L.W., Hamilton S.K., Peterson B.J.
1197 (2004). Stream denitrification and total nitrate uptake measured using a ¹⁵N tracer addition
1198 approach. *Limnology and Oceanography* 49, 809–820.

1199 Muñoz, I., Real, M., Guasch, H., Navarro, E., Sabater, S. (2001). Effects of atrazine on periphyton
1200 under grazing pressure. *Aquatic Toxicology* 55, 239-249.

1201 Murdock, J.N., Shields, F.D., Lizotte, R.E. (2013). Periphyton responses to nutrient and atrazine
1202 mixtures introduced through agricultural runoff. *Ecotoxicology* 22, 215–230.

1203 Neury-Ormanni, J., Doose, C., Majdi, N., Vedrenne, J., Traunspurger, W., Morin, S., (2020). Selective
1204 grazing behaviour of chironomids on microalgae under pesticide pressure. *Science of the*
1205 *Total Environment* 730, 1–8.

1206 Newman, M.C., Clements, W.H. (2008) Experimental approaches in community ecology and
1207 ecotoxicology. In: *Ecotoxicology: A comprehensive treatment* (Newman, M. C. Ed.), pp 439-
1208 467, Taylor and Francis, Boca Raton, FL.

1209 Page, M. J., McKenzie, J. E., Bossuyt, P. M., Boutron, I., Hoffmann, T. C., Mulrow, C. D. , Shamseer, L.,
1210 Tetzlaff, J. M., Akl, E. A., Brennan, S. E., R. Chou, J. Glanville, J. M. Grimshaw, A. Hróbjartsson,
1211 M. M. Lalu, T. Li, E. W. Loder, E. Mayo-Wilson, S. McDonald, L. A. McGuinness, L. A. Stewart,
1212 J. Thomas, A. C. Tricco, V. A. Welch, P. Whiting, Moher, D. (2021). The PRISMA 2020
1213 statement: an updated guideline for reporting systematic reviews. *BMJ* 372, n71.

1214 Paule, A., Biaz, A., Leflaive, J., Lawrence, J.R., Rols, J.L. (2015a). Fate of the Herbicide Alachlor
1215 Exposed to Different Microbial Consortia in Aquatic Systems. *Water Air and Soil Pollution*
1216 226, 3.

1217 Paule, A., Lamy, A., Roubéix, V., Delmas, F., Rols, J.L. (2015b). Influence of the natural growth
1218 environment on the sensitivity of phototrophic biofilm to herbicide. *Environmental Science*
1219 *and Pollution Research* 22, 8031-8043.

1220 Pazos, M., Ferreira, L., Rosales, E., Sanroman, M.A. (2016). Biofilm-mediated Degradation of
1221 Polycyclic Aromatic Hydrocarbons and Pesticides. In: *Biofilms in Bioremediation: Current*
1222 *Research and Emerging Technologies* (Edited by: G. Lear). Caister Academic Press, U.K.

1223 Pérez, G.L., Torremorell, A., Mugni, H., Rodríguez, P., Solange Vera, M., do Nascimento, M., Allende,
1224 L., Bustingorry, J., Escaray, R., Ferraro, M., Izaguirre, I., Pizarro, H., Bonetto, C., Morris, D.P.,
1225 Zagarese, H. (2007). Effects of the herbicide Roundup on freshwater microbial communities:
1226 a mesocosm study. *Ecological Applications* 17, 2310-2322.

1227 Pesce, S., Mamy, L., Achard, A. L., Le Gall, M., Le Perchec, S., Rechauchère, O., ... Sanchez, W. (2021).
1228 Collective scientific assessment as a relevant tool to inform public debate and policymaking:
1229 an illustration about the effects of plant protection products on biodiversity and ecosystem
1230 services. *Environmental Science and Pollution Research* 28, 38448-38454.

1231 Pesce, S., Zoghlami, O., Margoum, C., Artigas, J., Chaumot, A., Foulquier, A. (2016). Combined effects
1232 of drought and the fungicide tebuconazole on aquatic leaf litter decomposition. *Aquatic*
1233 *Toxicology* 173, 120-131.

1234 Pesce, S., Bouchez, A., Montuelle, B. (2011). Effects of organic herbicides on phototrophic microbial
1235 communities in freshwater ecosystems. *Reviews of Environmental Contamination and*
1236 *Toxicology* 214, 87-124.

1237 Pesce, S., Margoum, C., & Montuelle, B. (2010). In situ relationships between spatio-temporal
1238 variations in diuron concentrations and phototrophic biofilm tolerance in a contaminated
1239 river. *Water Research*, 44, 1941-1949.

1240 Pesce, S., Batisson, I., Bardot, C., Fajon, C., Portelli, C., Montuelle, B., Bohatier, J. (2009). Response of
1241 spring and summer riverine microbial communities following glyphosate exposure.
1242 *Ecotoxicology and Environmental Safety* 72, 1905-1912.

- 1243 Pesce, S., Fajon, C., Bardot, C., Bonnemoy, F., Portelli, C., Bohatier, J. (2008). Longitudinal changes in
1244 microbial planktonic communities of a French river in relation to pesticide and nutrient
1245 inputs. *Aquatic Toxicology* 86, 352-360.
- 1246 Pesce, S., Fajon, C., Bardot, C., Bonnemoy, F., Portelli, C., Bohatier, J. (2006). Effects of the
1247 phenylurea herbicide diuron on natural riverine microbial communities in an experimental
1248 study. *Aquatic Toxicology* 78, 303-314.
- 1249 Pettorelli, N., Schulte to Bühne, H., Tulloch, A., Dubois, G., Macinnis-Ng, C., Queirós, A. M., ... &
1250 Nicholson, E. (2018). Satellite remote sensing of ecosystem functions: opportunities,
1251 challenges and way forward. *Remote Sensing in Ecology and Conservation*, 4, 71-93.
- 1252 Pileggi, M., Pileggi, S. A., & Sadowsky, M. J. (2020). Herbicide bioremediation: from strains to
1253 bacterial communities. *Heliyon*, 6, e05767.
- 1254 Pimentao, A.R., Pascoal, C., Castro, B.B., Cassio, F. (2020). Fungistatic effect of agrochemical and
1255 pharmaceutical fungicides on non-target aquatic decomposers does not translate into
1256 decreased fungi- or invertebrate-mediated decomposition. *Science of the Total Environment*
1257 712, 10.
- 1258 Pinckney, J.L., Ornofsdottir, E.B., Lumsden, S.E. (2002). Estuarine phytoplankton group-specific
1259 responses to sublethal concentrations of the agricultural herbicide, atrazine. *Marine*
1260 *Pollution Bulletin* 44, 1109-1116.
- 1261 Piscart, C., Navel, S., Maazouzi, C., Montuelle, B., Cornut, J., Mermillod-Blondin, F., ... Marmonier, P.
1262 (2011). Leaf litter recycling in benthic and hyporheic layers in agricultural streams with
1263 different types of land use. *Science of the Total Environment* 409, 4373-4380.
- 1264 Polst, B.H., Anlanger, C., Risse-Buhl, U., Larras, F., Hein, T., Weitere, M., Schmitt-Jansen, M. (2018).
1265 Hydrodynamics Alter the Tolerance of Autotrophic Biofilm Communities Toward Herbicides.
1266 *Frontiers in Microbiology* 9, 2884
- 1267 Preston-Mafham, J., Boddy, L., Randerson, P. F. (2002). Analysis of microbial community functional
1268 diversity using sole-carbon-source utilisation profiles—a critique. *FEMS Microbiology Ecology*
1269 42, 1-14.
- 1270 Pringault, O., Lafabrie, C., Avezac, M., Bancon-Montigny, C., Carre, C., Chalghaf, M., Delpoux, S.,
1271 Duvivier, A., Elbaz-Poulichet, F., Gonzalez, C., Got, P., Leboulanger, C., Spinelli, S., Hlaili, A.S.,
1272 Bouvy, M. (2016). Consequences of contaminant mixture on the dynamics and functional
1273 diversity of bacterioplankton in a southwestern Mediterranean coastal ecosystem.
1274 *Chemosphere* 144, 1060-1073.
- 1275 Proia, L., Cassio, F., Pascoal, C., Tlili, A., Romani, A.M., Geislinger, A., Guasch, H. (2012). The use of
1276 attached microbial communities to assess ecological risks of pollutants in freshwater
1277 ecosystems. The role of heterotrophs. *Emerging and Priority Pollutants in Rivers: Bringing*
1278 *science into River Management Plans*, Springer, 55-83.
- 1279 Proia, L., Morin, S., Peipoch, M., Romani, A.M., Sabater, S. (2011). Resistance and recovery of river
1280 biofilms receiving short pulses of Triclosan and Diuron. *Science of the Total Environment* 409,
1281 3129-3137.
- 1282 Qiu, Y.W., Zeng, E.Y., Qiu, H.L., Yu, K.F., Cai, S.Q. (2017). Bioconcentration of polybrominated
1283 diphenyl ethers and organochlorine pesticides in algae is an important contaminant route to
1284 higher trophic levels. *Science of the Total Environment* 579, 1885-1893.
- 1285 Ralph, P.J., Smith, R.A., Macinnis-Ng, C.M.O., Seery, C.R. (2007). Use of fluorescence-based
1286 ecotoxicological bioassays in monitoring toxicants and pollution in aquatic systems: Review.
1287 *Toxicological & Environmental Chemistry* 89, 589-607.

1288 Rand, G.M., Clark, J.R., Holmes, C.M. (2001). The use of outdoor freshwater pond microcosms. III.
1289 Responses of phytoplankton and periphyton to pyridaben. *Environmental Toxicology* 16, 96-
1290 103.

1291 Rasmussen, J.J., Wiberg-Larsen, P., Baattrup-Pedersen, A., Monberg, R.J., Kronvang, B. (2012).
1292 Impacts of pesticides and natural stressors on leaf litter decomposition in agricultural
1293 streams. *Science of the Total Environment* 416, 148-155.

1294 Ricart, M., Barcelo, D., Geiszinger, A., Guasch, H., de Alda, M.L., Romani, A.M., Vidal, G., Villagrasa,
1295 M., Sabater, S. (2009). Effects of low concentrations of the phenylurea herbicide diuron on
1296 biofilm algae and bacteria. *Chemosphere* 76, 1392-1401.

1297 Rimet, F., Bouchez, A. (2011). Use of diatom life-forms and ecological guilds to assess pesticide
1298 contamination in rivers: Lotic mesocosm approaches. *Ecological Indicators* 11, 489-499.

1299 Rohr, J.R., Crumrine, P.W. (2005). Effects of an herbicide and an insecticide on pond community
1300 structure. *Ecological Applications* 15, 1135-1147.

1301 Rooney, R.C., Davy, C., Gilbert, J., Prosser, R., Robichaud, C., Sheedy, C. (2020). Periphyton
1302 bioconcentrates pesticides downstream of catchment dominated by agricultural land use.
1303 *Science of the Total Environment* 702, 134472.

1304 Rossi, F., Mallet, C., Portelli, C., Donnadiou, F., Bonnemoy, F., Artigas, J. (2019). Stimulation or
1305 inhibition: Leaf microbial decomposition in streams subjected to complex chemical
1306 contamination. *Science of the Total Environment* 648, 1371-1383.

1307 Rossi, F., Pesce, S., Mallet, C., Margoum, C., Chaumot, A., Masson, M., Artigas, J. (2018). Interactive
1308 Effects of Pesticides and Nutrients on Microbial Communities Responsible of Litter
1309 Decomposition in Streams. *Frontiers in Microbiology* 9, 13.

1310 Roubex, V., Pesce, S., Mazzella, N., Coste, M., Delmas, F. (2012). Variations in periphytic diatom
1311 tolerance to agricultural pesticides in a contaminated river: An analysis at different diversity
1312 levels. *Fresenius Environmental Bulletin* 21, 2090-2094.

1313 Roubex, V., Mazzella, N., Mechin, B., Coste, M., Delmas, F. (2011). Impact of the herbicide
1314 metolachlor on river periphytic diatoms: Experimental comparison of descriptors at different
1315 biological organization levels. *Annales de Limnologie-International Journal of Limnology* 47,
1316 239–249.

1317 Sabater, S., Guasch, H., Ricart, M., Romani, A., Vidal, G., Klünder, C., Schmitt-Jansen, M. (2007).
1318 Monitoring the effect of chemicals on biological communities. The biofilm as an interface.
1319 *Analytical and Bioanalytical Chemistry* 387, 1425-1434.

1320 Saxton, M.A., Morrow, E.A., Bourbonniere, R.A., Wilhelm, S.W. (2011). Glyphosate influence on
1321 phytoplankton community structure in Lake Erie. *Journal of Great Lakes Research* 37, 683-
1322 690.

1323 Schaffer, J.D., Sebetich, M.J. (2004). Effects of aquatic herbicides on primary productivity of
1324 phytoplankton in the laboratory. *Bulletin of Environmental Contamination and Toxicology* 72,
1325 1032-1037.

1326 Schmitt-Jansen, M., Altenburger, R. (2005). Toxic effects of isoproturon on periphyton communities -
1327 a microcosm study. *Estuarine Coastal and Shelf Science* 62, 304-312.

1328 Sharma, A., Kumar, V., Shahzad, B. et al. (2019). Worldwide pesticide usage and its impacts on
1329 ecosystem. *SN Applied Sciences* 1, 1446.

1330 Sheng, Y., Benmati, M., Guendouzi, S., Benmati, H., Yuan, Y., Song, J., Xia, C., Berkani, M. (2022).
1331 Latest eco-friendly approaches for pesticides decontamination using microorganisms and
1332 consortia microalgae: A comprehensive insight, challenges, and perspectives. *Chemosphere*
1333 308, 136183.

- 1334 Smedbol, É., Gomes, M.P., Paquet, S., Labrecque, M., Lepage, L., Lucotte, M., Juneau, P. (2018).
1335 Effects of low concentrations of glyphosate-based herbicide factor 540® on an agricultural
1336 stream freshwater phytoplankton community. *Chemosphere* 192, 133-141.
- 1337 Staley, Z. R., Harwood, V. J., & Rohr, J. R. (2015). A synthesis of the effects of pesticides on microbial
1338 persistence in aquatic ecosystems. *Critical reviews in toxicology*, 45, 813-836.
- 1339 Su, X.X., Chen, Y., Wang, Y.Y., Yang, X.Y., He, Q. (2019). Impacts of chlorothalonil on denitrification
1340 and N₂O emission in riparian sediments: Microbial metabolism mechanism. *Water Research*
1341 148, 188-197.
- 1342 Sun, F. L., Wang, Y. S., Sun, C. C., Peng, Y. L., Deng, C. (2012). Effects of three different PAHs on
1343 nitrogen-fixing bacterial diversity in mangrove sediment. *Ecotoxicology* 21, 1651-1660.
- 1344 Sura, S., Waiser, M.J., Tumber, V., Raina-Fulton, R., Cessna, A.J. (2015). Effects of a herbicide mixture
1345 on primary and bacterial productivity in four prairie wetlands with varying salinities: An
1346 enclosure approach. *Science of the Total Environment* 512, 526-539.
- 1347 Sura, S., Waiser, M., Tumber, V., Farenhorst, A. (2012). Effects of herbicide mixture on microbial
1348 communities in prairie wetland ecosystems: A whole wetland approach. *Science of the Total*
1349 *Environment* 435, 34-43.
- 1350 Sutherland, D.L., Ralph, P.J. (2019). Microalgal bioremediation of emerging contaminants -
1351 Opportunities and challenges. *Water Research* 164, 114921.
- 1352 Thomas, K.A., Hand, L.H. (2011). Assessing the potential for algae and macrophytes to degrade crop
1353 protection products in aquatic ecosystems. *Environmental Toxicology and Chemistry* 30, 622-
1354 631.
- 1355 Tien, C.J., Lin, M.C., Chiu, W.H., Chen, C.S. (2013). Biodegradation of carbamate pesticides by natural
1356 river biofilms in different seasons and their effects on biofilm community
1357 structure. *Environmental Pollution* 179, 95-104.
- 1358 Tlili, A., Berard, A., Blanck, H., Bouchez, A., Cássio, F., Eriksson, K. M., ... Behra, R. (2016).
1359 Pollution-induced community tolerance (PICT): towards an ecologically relevant risk
1360 assessment of chemicals in aquatic systems. *Freshwater Biology* 61, 2141-2151.
- 1361 Tlili, A., Montuelle, B., Berard, A., Bouchez, A. (2011). Impact of chronic and acute pesticide
1362 exposures on periphyton communities. *Science of the Total Environment* 409, 2102-2113.
- 1363 Tlili, A., Marechal, M., Montuelle, B., Volat, B., Dorigo, U., & Bérard, A. (2011). Use of the MicroResp™
1364 method to assess pollution-induced community tolerance to metals for lotic biofilms.
1365 *Environmental Pollution*, 159(1), 18-24.
- 1366 Vadeboncoeur, Y., Lodge, D. M., Carpenter, S. R. (2001). Whole-lake fertilization effects on
1367 distribution of primary production between benthic and pelagic habitats. *Ecology* 82, 1065-
1368 1077.
- 1369 Vera, M.S., Lagomarsino, L., Sylvester, M., Perez, G.L., Rodriguez, P., Mugni, H., Sinistro, R., Ferraro,
1370 M., Bonetto, C.; Zagarese, H., Pizarro, H. (2010). New evidences of Roundup (glyphosate
1371 formulation) impact on the periphyton community and the water quality of freshwater
1372 ecosystems. *Ecotoxicology* 19, 710-721.
- 1373 Vesterinen, J., Devlin, S. P., Syväranta, J., Jones, R. I. (2017). Influence of littoral periphyton on
1374 whole-lake metabolism relates to littoral vegetation in humic lakes. *Ecology* 98, 3074-3085.
- 1375 Vignati, D. A., Ferrari, B. J., Dominik, J. (2007). Laboratory-to-field extrapolation in aquatic
1376 sciences. *Environmental Science and Technology* 41, 1067-1073.
- 1377 Villeneuve, A., Montuelle, B., Bouchez, A. (2011). Effects of flow regime and pesticides on periphytic
1378 communities: Evolution and role of biodiversity. *Aquatic Toxicology* 102, 123-133.

1379 Vonk, J.A., Kraak, M.H.S. (2020). Herbicide Exposure and Toxicity to Aquatic Primary Producers.
1380 *Reviews of Environmental Contamination and Toxicology* 250, 119-171.

1381 Widenfalk, A., Svensson, J.M., Goedkoop, W. (2004). Effects of the pesticides captan, deltamethrin,
1382 isoproturon, and pirimicarb on the microbial community of a freshwater sediment.
1383 *Environmental Toxicology and Chemistry* 23, 1920-1927.

1384 Widenfalk, A., Bertilsson, S., Sundh, I., Goedkoop, W. (2008). Effects of pesticides on community
1385 composition and activity of sediment microbes - responses at various levels of microbial
1386 community organization. *Environmental Pollution* 152, 576-584.

1387 Wood, R.J., Mitrovic, S.M., Lim, R.P., Warne, M. St J., Dunlop, J., Kefford, B.J. (2019). Benthic diatoms
1388 as indicators of herbicide toxicity in rivers – A new SPEcies At Risk (SPEARherbicides) index.
1389 *Ecological Indicators* 99, 203-213.

1390 Wood, R.J., Mitrovic, S.M., Lim, R.P., Kefford, B.J. (2017). Chronic effects of atrazine exposure and
1391 recovery in freshwater benthic diatoms from two communities with different pollution
1392 histories. *Aquatic Toxicology* 189, 200-208.

1393 Zhang, M., Liu, W., Qu, Q., Ke, M., Zhang, Z., Zhou, Z., Lu, T., Qian, H. (2020). Metabolomic
1394 modulations in a freshwater microbial community exposed to the fungicide azoxystrobin.
1395 *Journal of Environmental Sciences* 97, 102-109.

1396 Zhao, Y., Zhang, Y., Guo, J. Wang, J., Li, Y. (2022). Shifts in periphyton research themes over the past
1397 three decades. *Environmental Science and Pollution Research*,
1398 <https://doi.org/10.1007/s11356-022-24251-7>

1399 Zhao, Z.H., Zhang, L., Deng, J.M., Wu, J.L. (2015). The potential effects of phytoplankton on the
1400 occurrence of organochlorine pesticides (OCPs) and polycyclic aromatic hydrocarbons (PAHs)
1401 in water from Lake Taihu, China. *Environmental Science-Processes & Impacts* 17, 1150-1156.

1402 Zubrod, J.P., Englert, D., Wolfram, J., Wallace, D., Schnetzer, N., Baudy, P., ... Bundschuh, M. (2015).
1403 Waterborne toxicity and diet-related effects of fungicides in the key leaf shredder *Gammarus*
1404 *fossarum* (Crustacea: Amphipoda). *Aquatic Toxicology* 169, 105-112.

1405 Zubrod, J.P., Bundschuh, M., Feckler, A., Englert, D., Schulz, R. (2011). Ecotoxicological impact of the
1406 fungicide tebuconazole on an aquatic decomposer-detritivore system. *Environmental*
1407 *Toxicology and Chemistry* 30, 2718-2724.