Wastewater quality and required water quality for irrigation purposes

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Report summary:

This review summarizes the main issues related to wastewater reuse for agricultural irrigation. It discusses successively the reuse itself, the sanitary, environmental and agricultural hazards caused by human pathogens and chemicals in wastewater, the economic sustainability of wastewater reuse, and the means either legal (guidelines or regulations and standards) or economic (tariffs, subsidies, taxes) to promote wastewater reuse while minimizing the risks. Proposals are made in the conclusion to improve and promote wastewater reuse in a more rational framework. As human enteric viruses are more and more often incriminated in human outbreaks, Appendix 1 provides a review on their fate in the environment.

Although wastewater reuse can address simultaneously problems in water quantity and quality, it remains low in most of the European countries. Wastewater is preferentially reused for crop irrigation in South European countries having a high Water Stress Index, high water needs for crops and large volumes of wastewater produced (Cyprus, Malta, Spain and Italy), and reuse will increase further in these countries. Wastewater reuse remains low in South European countries having a lower water stress index (Greece, France and Portugal), but it should increase because of global warming and the increase in frequency of extreme droughts. In more Northern European countries where water deficit for crops is lower or non-existent, wastewater may be reused locally for irrigation (e.g. in Germany) and/or in other sectors such as urban and industry sectors (e.g. in Belgium); several large cities and conurbations depend on recharging the surface and ground water bodies by treated wastewater. We have obtained nearly no information on wastewater reuse in Bulgaria that has one of the highest water stress index of European countries.

Public acceptance is good, but some opposition exists ("psychologically repugnant", "lack of purity", "can cause disease") and justifies information to prevent project failure. Actual risks include sanitary, environmental and agricultural hazards; they result from the presence in raw sewage of human pathogens and various inorganic and organic compounds. Although it is possible to produce water of almost any quality desired from wastewater, cost-effectiveness of treatments must be ensured. In order to protect conventional water resources, reduce the risks inherent in the use of wastewaters to tolerable levels, and insure the economic sustainability of reuse projects, the management of conventional and alternative water resources requires appropriate regulations and standards, as well as economic policy. Since water requirements, properties of raw wastewaters, and human resistance to pathogens vary with regions, it would not be appropriate to use the same regulations and standards in all European countries. However, the current diversity of rules is not scientifically justified and lead to inequalities. Europe could propose guidelines with maximum tolerated risks and a methodological framework to elaborate regional or national regulations and standards that account for local specificities. A distinction should then be performed between crops for local markets or for export.

New tools to support decisions such as computer programs are required. They have to account simultaneously for treatments, hazards, and cost-benefit considerations (with the monetary valuations of changes in human health, environment and crops), whereas existing tools only address part of the problem: they include models for quantitative microbial risk assessment, decision support systems for the configuration of wastewater treatment plants, and methods for the monetary valuation of the positive and negative changes in the environment. In addition, there are still gaps in knowledge and not enough data or parameters estimated correctly to ensure the reliability of these models. First, the fate of pollutants (including pathogens) is not enough known and described to properly inform quantitative microbial risk assessment; second, the monetary valuation of environmental changes is recent and probably requires a step back with, where possible, a comparison of methods; third, several studies have questioned the relevance of standards in selected microbial indicators and recent methodological developments suggest that the direct monitoring of some pathogens would be more appropriate in the next years.

Key words: wastewater, reuse, irrigation, hazards, enteric pathogens, chemical pollutants, regulations, guidelines, standards, economic sustainability, cost-effectiveness, yuck factor

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Introduction: wastewater recycling as a mean to cope problems in water quantity and quality

The world is facing increasing problems of limited water resources (Scheierling et al., 2010; Escobar, 2010; Anderson, 2006; Lazarova and Asano, 2005) illustrated by the water stress index (Bixio et al., 2006), the depletion of groundwater tables (Cao et al., 2013; Taylor et al., 2012; Frederick, 2006) and the increase in the Palmer drought severity index (Bates et al., 2008). They result from population growth – e.g. in North Africa (Qadir et al., 2010; Ouanouki et al., 2009), in Jordan (Alfarra et al., 2011) and in arid West States of the USA –, from Earth global warning as in South European Countries (Bixio et al., 2006) – e.g. in Greece (García-Ruiz et al., 2011), Italy (García-Ruiz et al., 2011; Van der Bruggen, 2010; Giungato et al., 2010) and Spain (Estrela and Vargas, 2012; García-Ruiz et al., 2011; Pedrero et al., 2010; Esteban and De Miguel, 2008) –, from increasing urbanization (Kennedy et al., 2012; Bates et al., 2008) – e.g. in Windhoek (Du Pisani, 2006), San Diego (Steirer and Thorsen, 2013) and Mexico City (Tortajada and Castelán, 2003) –, the main problem being then to have water available at the right place and the right time with the required quality (Angelakis et al., 1999), and from the increase of recreational uses (Sinclair et al., 2009; Hamilton et al., 2007b). Frederick (2006) estimated that about 10% of the world's agricultural food output depends on non-renewable groundwater supplies, with water tables falling a meter or more annually in parts of Mexico, India, China, the USA, and several other countries. **The world is facing simultaneously a decrease in the quality of conventional waters** (Van der Bruggen, 2010) subject to chemical and microbiological pollutions (Bradbury et al., 2013; Van der Bruggen, 2010; Craun et al., 2006; Tillaut et al., 2004; Cournot et al., 2001). The overexploitation of coastal aquifers has led to their salinization (Giungato et al., 2010). And the discharge of inadequately treated wastewater (Leverenz et al., 2011; Okoh et al., 2007), usually only after a secondary treatment in Europe (European Commission, 2011; Commission of the European Communities, 1998; CEC, 1991), but sometimes after a lighter treatment, see without treatment especially in several developing countries (Raschid-Sally and Jayakody, 2008; Ensink and van der Hoek, 2006; Downs et al., 1999), has led to contaminations of rivers and aquifers by mineral and organic chemicals (Leverenz et al., 2011), as well as human pathogens (helminth eggs, protozoa, bacteria and viruses) (Pachepsky et al, 2011; Servais et al., 2009). In the European Community, these problems may be increased by the imperfect compliance (European Commission, 2011) with the Council Directive 91/271/EEC (CEC, 1991) and by the increase of the populations of large cities – e.g. Milan (Mazzini et al., 2013) –. The poor quality of conventional water has led to human recreational or drinkable waterborne outbreaks (Craun et al. 2010; Sinclair et al., 2009; Reynolds et al., 2008; Craun et al., 2006; Calderon et al., 2005), to various environmental effects like decrease in the biodiversity of rivers, soil compaction, eutrophication (Thomas et al., 2010; WHO-EC, 2002), and it may affect other human activities – e.g. tourism –. **More than energy, the scarcity and quality of water will limit further growth of established economies and the development of new ones** (Van der Bruggen, 2010), as it is already the case for China (Yi et al., 2011).

Several actions to cope problems of water quantity and quality can be considered: an improved maintenance of drinkable water mains and sewers to reduce leakage and contaminations (Craun et al., 2010; Reynolds et al., 2008), the reduction of irrigation that often explains most of water consumption (Jiménez and Asano, 2008) through changes in land use and agricultural practices, the sustainable discharge of wastewater in the environment, and the transportation of conventional water over long distances as already performed – e.g. in the US (Fort and Nelson, 2012), Israel (Tal, 2006), Spain (Downward and Taylor, 2007), between the state of Johore in Malaysia and Singapore (King, 2011; Tortajada, 2006), and between Turkey and Cyprus with a 107 km long pipeline that will go undersea and that should be completed in 2014 –. **These actions may be completed by the reuse of wastewater** (Bixio et al., 2008) for irrigation (Angelakis and Durham, 2008), industrial purposes (Van der Bruggen, 2010; Levine and Asano, 2002), urban uses (Van der Bruggen, 2010), domestic uses like toilet flushing (Van der Bruggen, 2010; Radcliffe, 2004) and drinking (Leverenz and al., 2011; Du Pisani, 2006). At the Earth scale, Van der Bruggen (2010) estimates than 60%, 30% and 10% of all reuse applications are to be found in agricultural irrigation, in industries (especially cooling water and process water recycling), and in irrigation of parks, sport field or for groundwater recharge by percolation, respectively. These proportions are broadly consistent with the proportions of conventional water uses: e.g., Levine and Asano (2002) noted that industrial water use comprises about 25% of all worldwide water withdrawals. It is the objective of countries to reclaim all the collected wastewaters – e.g. Cyprus (Papaiacovou and Papatheodoulou, 2013; Fatta and Anayiotou, 2007; Fatta et al., 2005) and Israel (Brenner, 2012; Friedler, 2001) –. **If all wastewaters would be collected and reclaimed, their reuse could correspond to more than about 5-15% of the country water consumptions**. Wastewater reuse can be supplemented by the more expensive desalination of salt or brackish water (Ghaffour et al., 2013; Peñate and García-Rodríguez, 2012; Lattemann et al., 2011; Garud et al., 2011), the desalination of treated wastewaters by reverse osmosis being also used to decrease their salinity for irrigation – e.g. in Alicante wastewater treatment plant, Spain (Renault et al., 2013) –.

Although wastewater reuse in agricultural irrigation has been an old practice – e.g. in Ancient Egypt and China (Van der Bruggen, 2010; Angelakis and Koutsoyiannis, 2003) and in ancient Greece and the island of Crete (Lofrano and Brown, 2010; Angelakis et al., 2005) –, **it was not until the last quarter of the 20th century that water reuse appeared on the international agenda, at first in industrialized countries such as America and Europe** (Van der Bruggen, 2010). Lofrano and Brown (2010) proposed a review on the evolution of wastewater management through the ages with a special emphasis to its impacts on human health and environment. Initially, it was no less than direct reuse of wastewater without any treatment, apart from possible dilution (Van der Bruggen, 2010), as it is still the case on today in numerous places like Pakistan (Ensink and van der Hoek, 2006), Mexico (Dows et al., 2000, 1999), and in several developing countries (Raschid-Sally and Jayakody, 2008); and common practices exist in the developing world for families to reuse own wastewater, often for irrigation (Van der Bruggen, 2010). In the Middle Age in large cities of Europe, severe problems arose with surface water quality due to city population growth as rivers generally were used simultaneously as sewers and for water supply. Several cholera outbreaks were thus observed in London – e.g. in 1854 (Brody et al., 2000) –, Paris – e.g. in 1832 (Kudlick, 1999) –, and in other big cities. In Amsterdam, problems have been amplified after the construction of a dam on the Amstel River to prevent intrusion of saline water of the Flevo Lake, as the city's ring of canals was soon highly polluted by discharge of wastewater (Van der Bruggen, 2010). It was not until the 16th century that measures were taken (Van der Bruggen, 2010), which include the establishment of sewage farms to "purify" wastewater by infiltration into the soil (Crook et al., 2005; Asano, 2002): in Germany since about 1550 (Asano, 2006), the United Kingdom since 1700 (Asano, 2006), and in France near Paris in 1872 (Brissaud, 2002; Védry et al., 2001). Wastewater reuse in industry developed only during the 20th century (Levine and Asano, 2002). The dominant water-using industries include electric power generation, petroleum refining, and production of steel, paper, chemicals and allied products; water is used for processing, washing, cooling – almost two-thirds of all industrial water –, and/or transporting products or materials. Industrial reuse have been in place in the USA since the 1940s, with chlorinated domestic wastewater effluent used for steel processing, and in Japan in 1951 where the purified water of the Mikawashima Wastewater Treatment Plant in Tokyo was reused as process water for a paper mill (Levine and Asano, 2002). The rapid industrial growth in Japan in the 1970s resulted in competition for water between industry and agriculture for available water sources, and about 80% of industrial process water in Japan was reused in 2000 (Levine and Asano, 2002). In China, the 1989 average rate of industrial water reuse was reported to be about 56% among 82 major cities, with a maximum reuse of 93% (Levine and Asano, 2002). Progressively, wastewater was also reused as recreational water, as early in 1965, the Santee, California recreational lakes, supplied with reused wastewater, were opened for swimming (Asano, 1998). Since this time, there have been several other examples (Sinclair et al., 2009; Craun et al., 2005). Until today, there have been only a few projects of potable wastewater reuse (Gerrity et al., 2013; Rodriguez et al., 2009a). The most famous example of Direct Potable Reuse (DPR), i.e. theoretically pipe-to-pipe, is in Windhoek, Namibia, since 1969 with increment in 1997 (Du Pisani, 2006), but other projects are operational, – e.g. at Cloudcroft, New Mexico, a tiny mountain resort town that mingles reclaimed water with local well water (Leverenz and al., 2011) –, or are planned – e.g. at Big Springs, Texas, (Leverenz et al., 2011) –. The failure of the initial project in San Diego in 1998 illustrates the difficulties for the public to accept direct contact with wastewater, the water department’s initiative being derided at this time as "toilet to tap" (Staub et al., 2011). Progressive changes in San Diego (Steirer and Thorsen, 2013) resulted firstly from subsequent water shortages and rationing, from the water department's that began reaching out to customers with discussion groups and public meetings, from the awareness of inhabitants that conventional waters receive treated wastewaters, and from the choice of indirect potable reuse option with treated wastewater first stored in San Vicente Reservoir. Indirect potable reuse (IPR), i.e. after blending with conventional water and a long storage underground or in reservoirs, is more widespread and several projects are operational (Rodriguez et al., 2009a), one of the most famous project being the Water Factory 21 in Orange County West District, California (Leverenz et al., 2011; Van der Bruggen, 2010), but funnelling reclaimed water into water supplies is being considered in other cities like Miami, Florida, and Denver, Colorado; in the North of Virginia, reclaimed water has flowed into the Occoquan Reservoir for three decades (Rose et al., 2001). In Asia, the NEWater project is to reduce Singapore dependency to Malaysia and officials consider that 15% of water originates from treated effluent; most of the treated wastewater is for irrigation or manufacturing but a small proportion is introduced to raw water reservoirs since 2003 for indirect potable reuse (Van der Bruggen, 2010). In Belgium, indirect potable reuse is carried out by the local drinking company IWVA in the Torreele/St-André project to supply potable water to the cities of De Panne, Koksijde, Nieuwport, Veume and Alveringem (Van Houtte et al., 2012), and large cities, such as London and Berlin depend on recharging the surface and groundwater bodies by treated wastewater (Paul and Blunt, 2012; Rygaard et al., 2011; Angelakis, 2011). More generally, the use of wastewater for groundwater recharge (Levantesi et al., 2010; Foster and Chilton, 2004; Asano and Cotruvo, 2004; Brissaud, 2003) may indirectly contribute to potable water supply, even if it is firstly to fulfil other objectives. *De facto*, indirect potable reuse is occurring widely, as conventional water used to produce tap water has often the discharge of treated or untreated wastewater, but usually is not acknowledged. **Until now, the proportion of wastewater that is reused has remained generally low: at the Earth level, even agricultural reuse represents only less than 1% in volume of the total demand of water by this sector** (Jiménez and Asano, 2008; Wade Miller, 2006).

Wastewater reuse in irrigation may address simultaneously water quantity and quality problems (Qadir et al., 2010; Angelakis and Durham, 2008; Hamilton et al., 2007b; Wade Miller, 2006; Bixio et al., 2006; Anderson, 2006; Asano, 2002 Friedler, 2001; Angelakis et al., 1999; Asano and Levine, 1996, among others), as irrigation often explains most of country water consumption (Van der Bruggen, 2010) and pollutants may be degraded or retained by soils. In addition, wastewater may contain fertilizing compounds (Ensink and van der Hoek, 2006) that lead farmers sometimes prefer to use untreated sewage rather than treated wastewater in developing countries like China, Mexico, Peru, Egypt, Lebanon, Morocco, India and Vietnam (Jiménez et al., 2010; Sophocleous et al., 2009; Keraita et al., 2008), as more extensively explained for Pakistan (Ensink and van der Hoek, 2006) and Mexico (Scott et al., 2000; Downs et al., 2000; Downs et al., 1999). Numerous papers have proposed overviews on the state of the art on wastewater reuse for irrigation on the Earth (Hochstrat and EUREAU members, 2011; Van der Bruggen, 2010; Qadir et al., 2010b; EUREAU, 2009; Angelakis and Durham, 2008; Raschid-Sally and Jayakody, 2008; O'Connor et al., 2008; Jiménez and Asano, 2008; Bixio et al., 2008; Keraita et al., 2008; Hamilton et al., 2007b; MED WWR WG, 2007; Bixio et al., 2006; Toze, 2006; Bixio et al., 2005; Aoki et al., 2005; Massoud et al., 2003; Hussain et al., 2002; Angelakis and Bontoux, 2001; Angelakis et al., 1999). Unfortunately, the nature of available information greatly differs with regions, leading to difficulties in comparisons and clear reporting; e.g. large parts of Asia were not included in the survey established by Bixio et al. (2005). In addition, small projects that predominate in several countries, including most of the underdeveloped countries, are not considered in surveys, leading to data without statistical significance (Bixio et al., 2005). Large-scale projects are mostly used for landscape and agricultural irrigation, whereas small-scale projects often have urban, recreational, or environmental uses; many relatively small-scale projects can be found in Japan, in contrast to the USA where water reclamation is mainly dominated by medium- to large-scale projects (Bixio et al., 2005). **Nowadays, there are great differences among countries** regarding the proportion of wastewater collected in sewers (Raschid-Sally and Jayakody, 2008; Hamilton et al., 2007b), the existence and nature of a subsequent treatments (Jiménez and Asano, 2008; Keraita et al., 2008; Bixio et al., 2008; Hamilton et al., 2007b) – irrigation may use raw wastewater (Raschid-Sally, 2010; Ensink and van der Hoek, 2006; Downs et al., 1999), diluted wastewater without treatment (Raschid-Sally, 2010; Keraita et al., 2008) or treated wastewater, and treatments also greatly vary between pre-treatments and advanced tertiary treatments –, and the proportion of wastewater reused in irrigation (Hamilton et al., 2007b). **Water recycling in irrigation is particularly practiced in world regions suffering water scarcity, such as the Near East and Middle East, Mediterranean countries, Australia, the southwest USA, and densely populated regions like Japan** (Lazarova and Asano, 2013; Bdour et al., 2009; Keraita et al., 2008; Radcliffe, 2004). **It is also practised in regions with severe restrictions on disposal of treated wastewater effluents**, such as Florida (O'Connor et al., 2008). Bixio et al. (2005) identified large water reuse projects– defined as above 0.5 $Mm^3.y^{-1}$ reclaimed water for unrestricted use or 2.5 $Mm^3.y^{-1}$ for restricted use – in Japan (over 1800), the USA (over 800), the EU (over 200), Australia (over 450), and around 100 sites In the Mediterranean and Middle East area, whereas 50 sites were found in Latin America and 20 in sub-Saharan Africa. Most of Asia (including China in which wastewater reuse is important (Chang and Ma, 2012; Yi et al., 2011)) was not included in their survey. Lazarova and Asano (2013) proposed estimates of the annual amount of reclaimed wastewater of about 5410 and 2770 $Mm^3.y^{-1}$ for China and United States of America, respectively. Other estimates for China are slightly lower although of the same order of magnitude (Yi et al., 2011). **In Europe, wastewater reuse in irrigation is practised mainly in Cyprus** (nearly 100% of treated wastewater), **Malta** (with a target of 25% of treated wastewater (Mangion, 2012))**, Spain** (Iglesias et al., 2010; Iglesias Esteban and Ortega de Miguel, 2008)**, Italy** (Mangion, 2012) **and to a lesser extent in Greece** (Guardiola-Claramonte et al., 2012) **with Crete Island** (Agrafioti and Diamadopoulos, 2012), **France** (Guardiola-Claramonte et al., 2012) in inland, coastal, and island areas – e.g. in the Noirmoutier Island (Fazio et al., 2013))**, in Germany and some other countries** (Guardiola-Claramonte et al., 2012; Van der Bruggen, 2010; EUREAU, 2004). Wastewater reuse seems to be negligible in Bulgaria (Hochstrat et al., 2006), although it has one of the highest water stress indexes among European countries (i.e. over 60%) (Bixio et al., 2006), but we found nearly no information on reuse in this country. Belgium which has also a high water stress index (over 40%) reuses wastewater mainly for urban and industrial uses (Bixio et al., 2008). Near Europe, wastewater reuse in irrigation is important in Israel (Tal, 2006), in Turkey and in some Mediterranean countries of the Middle East and North Africa (Qadir et al., 2010; Keraita et al., 2008). Reclaimed wastewater may be used through drip irrigation (Capra and Scicolone, 2007), sprinkler irrigation, and flooding – e.g. for paddy rice fields as in the south of Valencia, Spain (Renault et al., 2013) –.

Strategies chosen for wastewater reuse are highly variable in terms of requirements for environmental protection and human health, as may be illustrated by the respective choices of California and Mexico. In the USA, southern states (Florida, Texas, New Mexico, Arizona, California, Colorado and Nevada) are the most active in water recycling, with California and Florida recycling approximately one half of USA recycled wastewater (Van der Bruggen, 2010; Crook et al., 2005). For most of the four decades beginning in 1970, the arid West was the fastest-growing region in the USA, e.g. the population of Nevada quintupled in that period while Arizona's nearly quadrupled, the continued population growth being unmatched by growth in water storage capacity. The situation is such that, in California, San Diego rainfalls meet only about 15% of the needs and most of the water is imported from the Colorado River and the Sacramento-San Joaquin River Delta (Tchobanoglous, 2012). Wastewater recycling projects in California have been thus booming, with ca. 600 $Mm^3.y^{-1}$ of recycled water being used across over 4800 locations from 234 wastewater treatment plants (Van der Bruggen, 2010). However, the development of programs for planned reuse of wastewater within the California began in the early part of the 20th century (Asano, 2006): by 1910, 35 California communities were already using sewer water for irrigation; the city of Bakersfield has used reclaimed water since 1912 to irrigate corn, barley, alfalfa, cotton, and pasture (Asano, 2006), and in 1929, the city of Pomona initiated a project using reclaimed wastewater for the domestic irrigation of lawns and gardens. In 1970, water reclamation was formally encouraged in the California State Water Code (Asano, 1998); public health laws were progressively developed, leading to the publication of the so-called Title 22 (State of California, 2000) and the Purple Book (State of California, 2001), which are a collection of guidelines, rules, and standards corresponding to a zero tolerance that have been used later elsewhere as basis for regulations and standards (Bixio et al., 2008). The largest volumes are used for agricultural irrigation, and thereafter for landscape irrigation (Crook et al., 2005) – the nearly opposite proportions being observed in Florida (Crook et al., 2005) –, other applications including industrial reuse, groundwater recharge (Asano and Cotruvo, 2004), seawater barrier, recreation and wildlife, and indirect potable reuse. Irrigation is mainly for corn, barley, alfalfa, cotton, and pasture (Van der Bruggen, 2010). By contrast in Mexico, only a small proportion of wastewater is treated before their reclaim and reuse (Keraita et al., 2008), and wastewater is recycled in irrigation and considered for its fertilizing value in Guanajuato (Keraita et al., 2008; Scott et al., 2000) and in the Mezquital Valley (Jiménez, 2005). Farmers generally prefer untreated wastewater rather than treated water due to its fertilizing value that enable them to increase their rents for between 135 to 780 $US\$.ha^{-1}.y^{-1}$ (Keraita et al., 2008; Jiménez, 2005; Scott et al., 2000). A study comparing vegetable production using freshwater and untreated wastewater in Haroonabad, Pakistan, found that the gross margins were significantly higher for wastewater (US$150 per hectare), because farmers spent less on chemical fertilizer and achieved higher yields (Jiménez et al., 2010; Hussain et al., 2002).

The awareness that water recycling is a possible partial or total answer to the growing water needs is increasing, all the more that in water-stressed regions, water conflicts are already appearing, although sometimes still hidden (Van der Bruggen, 2010).

For examples, the California State Water Code stated in 1970 that *"it is the intention of the Legislature that the State undertake all possible steps to encourage development of water reclamation facilities so that reclaimed water be available to help meet the growing water requirements of the State"* (Asano, 2006; Asano and Levine, 1996) and, in the U.S., a milestone event was the passage of the Federal Water Pollution Control Act in 1972 (later renamed 'the CleanWater Act') *"to restore and maintain the chemical, physical, and biological integrity of the Nation's waters"* with the ultimate goal of zero discharge of pollutants into navigable, fishable, and/or swimmable waters (Asano, 2006). The European Communities declared in 1991 that *"treated wastewater shall be reused whenever appropriate. Disposal routes shall minimize the adverse effects on the environment"* (CEC, 1991). **Water recycling is the only possible solution in several situations**, as in continental areas without exploitable underground water resources, like in Windhoek, Namibia (du Pisani, 2006), and the reuse of wastewater can also be a way to cope with the problems of water quality, the decrease in quality being one of the most important cause of the decrease in quantity (Lazarova and Azano, 2013) as in China, which is now facing very serious problems of water and soil pollution (Yi et al., 2011). **The proportion of wastewater that is reused has remained generally low until now** (Jiménez and Asano, 2008), but there is a significant potential for an increased utilisation of reclaimed wastewater in European countries, specifically in the Mediterranean region, as showed by simulations considering some scenarios of water availability and uses (Hochstrat et al., 2006).

T**hus, the objectives of the rest of this report are:**

1. **To identify key success factors, constraints and milestones** to develop wastewater reuse in Europe. Constraints include economic sustainability, public acceptance, and potential risks for human health, the environment, and agricultural productions. Regulations and standards can simultaneously limit these risks and encourage the use of wastewater, as well as the economic policy (water price, taxes and subsidies). However, inadequate regulations and standards may be additional constraints. Positive interactions between stakeholders, including leaders of farmer associations are barely mentioned in this report, although they are an important key success factor; it is developed in work package W3.2;
2. **to propose in the conclusion of this report a list of multi-criteria requirements for using treated wastewater in irrigation** by integrating national regulations, reuse water scheme management constraints, health and agronomic constraints, etc..

This reports mainly based on (i) a bibliographic review (with several papers issued from European projects like AQUAREC and MEDAMARE), (ii) European & National regulations (including those on non-European countries/states), (iii) Feedback of each partner personal experiences, and (iv) available results of the WP3.2.

This review has been complicated by inaccurate data and contradictions between recent papers; inaccuracies and contradictions deal with permitted uses, regulations and standards for irrigation, and surface areas irrigated with treated wastewaters for several countries. They have resulted from the significant and rapid changes in wastewater reuse in several European countries during the last decade, from the lack of recent reviews and data compilation at National level for most European countries and, sometimes, from the difficulty of access to national regulations and standards. Probably the most complete and actualized informations are for Spain (Iglesias et al., 2010; Iglesias Esteban and Ortega de Miguel, 2008); by contrast, we don't find recent National review of the state of the art in Italy since the 'old' reviews of Barbagallo et al. (2001) and Bonomo et al. (1999), although recent regional informations exist – e.g. for the south of Milan (Mazzini et al., 2013), other parts of the Po valley (Verlicchi et al., 2012), the Apulia (Giungato et al., 2010), and Sicilia (Cirelli et al., 2012) –. As examples on encountered errors: (1) the unauthorized aquifer recharge with wastewaters in Cyprus (paper in 2012), while effluent from Paphos wastewater treatment plant are entirely used for Ezousa aquifer recharge (Papaiacovou and Papatheodoulou, 2013); (2) over 4000 ha of agricultural fields irrigated with wastewater in Italy (paper in 2007), whereas more than 3000 ha are concerned in the South of Milan, to which one has to add irrigated surface areas at least in other part of the Pô valley, in Apulia, Emilia Romagna, Sicilia (Cirelli et al., 2012) and Sardania; (2) the recommendations of the Conseil Supérieur d'Hygiène Publique de France (CSHPF, 1991) as current French regulations (paper in 2013), whereas current regulations and standards are from 2010 (Ministère de la Santé et des Sports, 2010). We hope to have avoided these pitfalls, and we have often preferred remaining cautious about numerical values.

Hazards related to wastewater reuse for irrigation

Hazards related to wastewater reuse for irrigation include health/sanitary risks, environmental risks and agricultural risks. They result from wastewater content in pathogens and chemicals.

Raw wastewaters may contain human enteric pathogens, i.e. viruses (Symonds et al., 2009)**, bacteria** (Pachepsky et al, 2011)**, protozoa** (Tzipori and Widmer, 2008)**, and helminth eggs** (Gupta et al., 2009; Ensink and van der Hoek, 2006), that may resist to wastewater treatments (Gupta et al., 2009; Ryu et al., 2007; Gerba and Smith, 2005; Gerba, 1999; Amahmid et al., 1999). Wastewater treatment plant workers may be infected as illustrated by antibodies against hepatitis A and/or hepatitis B detected in a significantly higher proportion of workers than of control population in Thessaloniki, Greece (Arvanitidou et al., 2004), although contrasted results have been found in Naples (Montuori et al., 2009). Pathogens have caused drinking and recreational waterborne outbreaks in developed countries like U.S. (Cann et al., 2013; Craun et al., 2012; Sinclair et al., 2009; Reynolds et al., 2008; Craun et al., 2006; Maunula et al., 2005; Calderon et al., 2005; Craun et al., 2005) and Europe (Lopman et al., 2003; Koopmans et al., 2000), and they can contaminate agricultural products during irrigation, even with conventional waters (Pachepsky et al., 2011). In addition to enteric pathogens, **increased temperature and stagnant water may favour the growth of human non-enteric water-based pathogens**, like *Legionella*, *Mycobacterium* and *Naegleria fowleri* (Cann et al., 2013; Reynolds et al., 2008).

Raw wastewaters contain also various mineral chemicals (Toze, 2006b; Unkovich et al., 2006) **and organic pollutants** (Toze, 2006b; Toze, 2006a). High contents in sodium cation have detrimental effects on the structure of soils (Mazzini et al., 2013), and high concentrations in sodium, chloride and borate ions are toxic for plants (Unkovich et al., 2006). Generally, heavy metals are of little concern for irrigation with treated wastewaters, unless wastewater is of industrial origin and/or is not sufficiently treated (Toze, 2006b). **Organic pollutants include chemicals of personal care products, various pharmaceutical products** (Nikolaou et al., 2007; Hernando et al., 2006)**, bisphenol A and phthalates** (Barnabé et al., 2008; Clara et al., 2010; Dargnat et al., 2009). When a unique sewer network collects both house wastewaters and runoff waters, **wastewaters may also contain pesticides** (Gasperi et al., 2008b; Blanchoud et al., 2007) **and polycyclic aromatic hydrocarbons** (Palmquist and Hanaeus, 2005). The removal efficiency of wastewater treatment plants varies with compounds and treatment trains (Watkinson et al., 2007), and several studies noted the presence of organic contaminants in the environment – e.g. antibiotic in watershed (Watkinson et al., 2009) –. **Solvents are produced during disinfection by chlorine** (Kim et al., 2003) **and toxins may be synthesized by cyanobacteria** whose blooms are favoured by wastewater, especially in ponds (Furtado et al., 2009; Barrington and Chadouani, 2008; Saqrane et al., 2008; Gehringer et al., 2003; Sivonen and Jones, 1998). Impacts on human health remain uncertain for irrigation reuse. They may *a priori* result from pharmaceutically active compounds and a large number of compounds that are known or suspected endocrine disruptors (Toze, 2006b). **Concentrations of most organic pollutants in urban raw or treated wastewaters are generally below the toxic level**s for humans, but potential **problems may result from the combined effects of several pollutants and/or their cumulative consequences over long-term periods**.

The use of untreated wastewater has led to local or regional disastrous effects, such as in the Po valley the loss of permeability and the contamination of soils as well as a decrease in the biodiversity of surface water bodies, and 30% of the pollution in the northern portion of the Adriatic Sea south of Venice before the implantation Nosedo and San Rocco wastewater treatment plants in the south of Milan (Mazzini et al., 2013). By contrast, **the success of several existing projects of treated wastewater reuse in irrigation has comforted the opinion that wastewater recycling may be considered as a 'zero-risk' practice**, when properly treated (Lazarova and Asano, 2013). However, **there are neither enough experimental evidences** (including epidemiological studies) **nor enough works on relevant processes to know whether wastewater management in reclaiming projects in Europe is sufficient** to prevent health risks. First, **it is often a challenge to attribute disease outbreaks to specific exposure routes** (i.e. foodborne, waterborne, airborne or through nearly direct faecal to oral way (Todd et al., 2008)) due to other contributing factors, especially in developing countries with a poor hygiene, sanitation and reduced access to safe drinking water (Drechsel et al., 2012), and numerous case of diseases are not recorded (Reynolds et al., 2008). Second, **the exposure of sensible populations to human enteric pathogens may be fatal for them**, these populations representing more than 25% of the humans in countries like the U.S. (Reynolds et al., 2008). Third, **importations of agricultural products from developing countries increase the risks** because of irrigation with water generally of lower quality (Hunter et al., 2009) and because microorganisms that participate to the normal intestinal flora of local populations like several *Escherichia* coli may have a pathogenic effect on foreign populations. Fourth, **there are new emerging microbial and chemical contaminations** – e.g. hepatitis E virus (Koopmans and Duizer, 2004), severe acute respiratory syndrome (SARS) which moves from the bat population to other animals and humans (Gundy et al., 2009; Bennett, 2006), multi antibioresistant bacteria like *Escherichia coli* (Cantón et al., 2008), emergent virus strains (La Rosa et al., 2012; Yen et al., 2011), endocrine disruptors and pharmaceutical compounds including antibiotics (Leverenz et al., 2011; Barnes et al., 2008; Al-Rifai et al., 2007; Karthikeyan and Meyer, 2006), solvents generated by chlorine oxidation treatments (Lee and von Gunten, 2010; Kim et al., 2003) –.

It may be distinguished between upstream technological risks related to wastewater treatment, distribution and quality changes between the outlet of wastewater treatment plant and the point of use, from downstream hazards related to undesirable effects of reuse on human health, environment and agriculture (**Figure 1**).

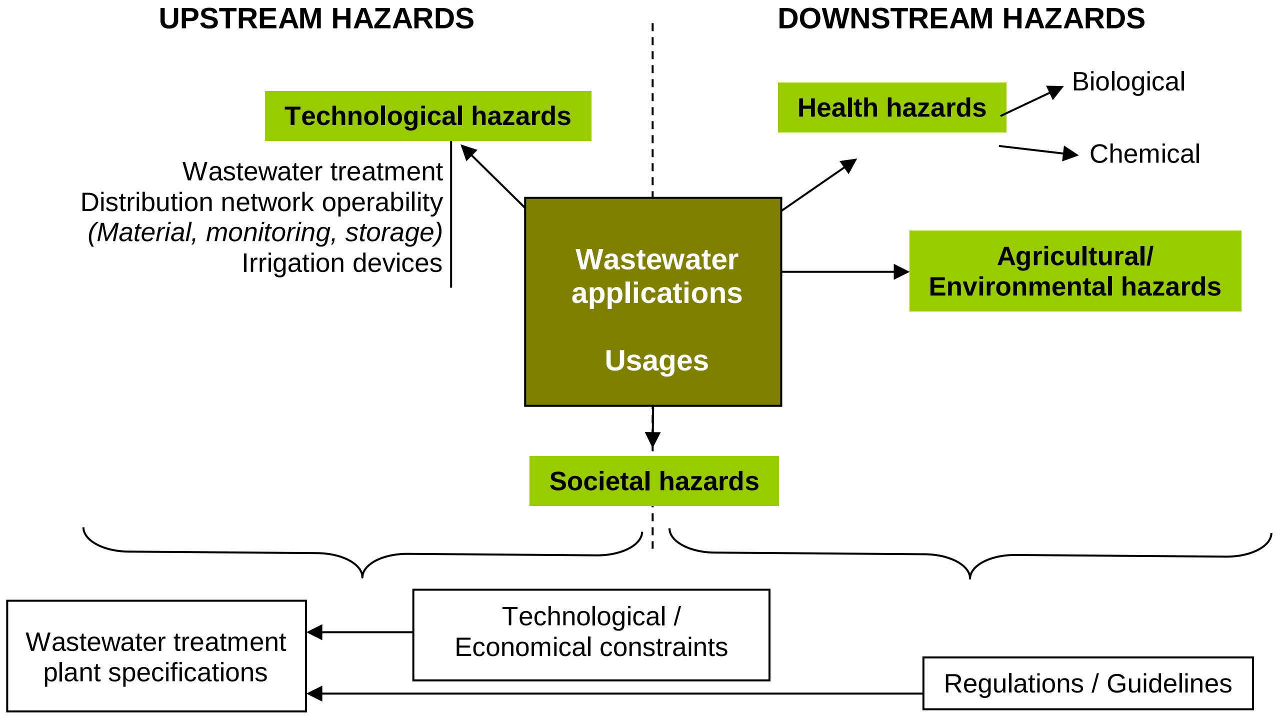

Figure 1: Hazards linked with wastewater reclaiming in irrigation.

Additional risks deal with the economic sustainability of wastewater reuse, and the acceptance or rejection of wastewater reclaiming by inhabitants (Leverenz et al., 2011; Cain, 2011; SOFRES, 2006). Even for direct potable water reuse, public acceptance may be good as in Windhoek, Namibia, (du Pisani, 2006); the failure of direct potable reuse in San Diego, California, (Staub et al., 2011) is almost fixed now thanks to several changes (Steirer and Thorsen, 2013; Shipps, 2013): inhabitants accept wastewater reuse all the more water deficit is an actual threat, water managers greatly communicate with consumers, and it becomes clear that recycled water is not a direct 'toilet to tap' transfer but that treatments insure a water of quality often higher than that of conventional waters and indirect potable reuse is now proposed. We will not discuss social acceptance further in this report, but it cannot be overlooked.

Technological hazards

Technological hazards include risks of inadequate wastewater treatments, of cross contamination between potable water mains and treated wastewater distribution network, of development of facultative enteric human pathogens and water-based pathogens in this network, and of biofilm with clogging hazards.

Today, **technically proven wastewater treatment and water purification processes exist to produce water of almost any quality desired**, and the quality of reclaimed water can exceed conventional drinking water quality based on most conventional parameters (Norton-Brandão et al., 2013; Asano, 2006; Asano, 1998). Efficient treatments are some tertiary ones, including physicochemical coagulation/flocculation, filtrations – especially ultra or nano-filtration (Pierre et al., 2010; Röhricht et al., 2010) including membrane bioreactor (Sima et al., 2011), and reverse osmosis (Peñate and García-Rodríguez, 2012; Garud et al, 2011), as sand filtration may be not sufficient for viruses (CFPTEP, 2010a; CFPTEP, 2010b) although some results are correct (Aronino et al., 2009) –, high UV radiation levels (Simonet and Gantzer, 2006b) or long stays in shallow lagoons that may lead to similar results, and chemical disinfection – e.g. with ozone (Martínez et al., 2011), hydrogen peroxide (Barrington and Chadouani, 2008), peracetic acid (Kitis, 2004), and chlorine or hypochlorite although they may lead to the production of solvent as by-products (Kim et al., 2003) –. However **in the European context, current treatments don't totally eliminate microbial and chemical pollutions due to costs**; and progress have still to be made to identify robust new methods of purifying water at lower cost and with less energy, while at the same time minimizing the use of chemicals and impact on the environment (Shannon et al., 2008). Several papers have reported large quantities of some human enteric viruses in raw and treated wastewaters – e.g. for the 5 wastewater treatment plants around Roma (La Rosa et al., 2010), in a wastewater treatment plant in the south of the Netherlands (Van den Berg et al., 2005), and for the Leipzig wastewater treatment plant (Pusch et al., 2005).

Failures in treatments may result from their lack, their inadequacy for specific microbial or chemical contaminants, or **their deficiency caused by a dysfunction of the plant or by temporary water overflows**. Municipal water systems can be overburdened by extreme rainfall events, as sewer systems often carry both storm water and wastewater (Cann et al., 2013; Figueras and Borrego, 2010). Similar reasons and the discharge of the excess wastewater directly into surface water bodies explain correlations between extreme rainfall events and waterborne disease outbreaks from drinking water (Cann et al., 2013). Although non-compliant treated wastewater should not be used, the frequency of water analyses and the waiting time for results make it impossible to react in real time to failures of quality. Treated wastewater may be stored in reservoirs or lagoons before irrigation. The storage is often regarded as an additional treatment enabling to break organic contaminants difficult to degrade in generally approximately 1 day of transit in the wastewater treatment plant. Especially for lagoons or reservoirs of small depth, solar UV radiations may accelerate some transformations, including the alteration of virus RNA or DNA (Fong and Lipp, 2005; Gerba et al., 2002; Fujioka and Yoneyama, 2002). Unfortunately, l**agoons or reservoirs may be contaminated by some of the human enteric pathogens** as *Salmonella enteritidis* (typhoid, diarrhea) and *Campylobacter jejuni* (diarrhea) from birds and *Cryptosporidum* (diarrhea) from cattle (Reynolds et al., 2008); in addition, wastewater favours blooms of cyanobacteria (Barrington and Chadouani, 2008) synthesizing neurotoxins and carcinogenic hepatotoxins (Sivonen and Jones, 1998) and, in several countries, water coming from ponds without special treatment contains microcystins that have an impact both on the growth and the development of crops (Furtado et al., 2009; Gehringer et al., 2003; Saqrane et al., 2008).

Leaks in treated wastewater distribution and sometimes interconnections with conventional water distribution systems, especially when using a unique pipe network of irrigation alternatively supplied with treated wastewater and conventional water, **may led to contaminations** of conventional water (Reynolds et al., 2008) and/or the environments (McKay, 2011). Groundwater contamination may also originate from household wells (Borchardt et al., 2003). Available statistics on potable water mains may probably be partly transposed to treated water distribution systems, especially in the U.S. even well-run water distribution systems experience about 25-30 breaks per 160 km of piping per year and the percent of leakage ranges between <10% to as high as 32% (Reynolds et al., 2008). Reynolds et al. (2008) also noted that at least 20% of the potable water mains is below the water table; probably the proportion is lower for treated wastewater distribution pipes, but leaks below the water table is probably the worst situation in terms of contamination of groundwater and/or drinking water in urban context. **Possible growth in pipe networks of human enteric pathogens** not totally eliminated during wastewater treatments **deals with some pathogens,** including *Escherichia* coli (Pachepsky et al., 2012), as all human enteric viruses, most human enteric protozoa and several bacteria are obligatory parasites. **In addition, increased temperature and stagnant water favour the growth of human water-based pathogens, like *Legionella*** (pneumonia, respiratory infections)**, *Mycobacterium*** (however, some *Mycobacterium*, including the tuberculosis and the leprosy organisms, are obligate parasites and are not found as free-living members of the genus) **and *Naegleria fowleri*** (causing primary amoebic meningoencephalitis (PAM), fortunately rare, of primary concern because of high fatality rate in diagnosed cases (>95%)) (Cann et al., 2013; Reynolds et al., 2008). If microbial biofilms seems not lead to clog pipes, **biofouling has been identified as a major contributor to emitter clogging in drip irrigation systems using reclaimed wastewater** (Pachepsky et al., 2012; Capra and Scicolone, 2007); it lead to proposal the structure of the emitters (Yan et al., 2009).However in some contexts, drip emitter clogging may be higher for irrigation with conventional water than with reclaimed water (Cirelli et al., 2012)..

Other technological hazards exist, including those intended to the internal reuse of grey waters for garden irrigation, and the reclaiming of industrial wastewaters, especially for many low-income countries with challenges of emerging industrial sectors or mining activities while institutional, technical and/or regulatory capacities for wastewater treatment are not yet in place; industrial effluents pose a threat to humans and the environment (Simmons et al., 2010).

Health/sanitary hazards

Numerous factors affect sanitary/health hazards. They depend on **the domestic and/or industrial origins of wastewater** that induce first microbial risks or chemical risks, respectively, although chemical risks may also be induced by domestic waters – e.g. antibiotics in Cyprus conventional waters (Papaiacovou and Papatheodoulou, 2013) –. They also depend on **the local importance of pathogens** – e.g. hepatitis A is endemic in North Africa countries (Kamal et al., 2010; Gharbi-Khelifi et al., 2006) and in some European countries like Albania (Divizia et al., 2005), widespread in some other European countries like Greece (Arvanitidou et al., 2004), and nearly absent in most of the other European countries except after contaminations due to international exchanges (Hollinger et Emerson, 2007); helminth eggs may prevail in Pakistan (Ensink and van der Hoek, 2006) and India (Gupta et al., 2009) –. They also depend on **seasonal variations** of some infections (Lal et al., 2012; Jagai et al., 2012; Yen et al., 2011; Gharbi-Khelifi et al., 2006; Chikhi-Brachet et al., 2002) although this seasonality is not necessarily found for virus in raw and treated wastewaters – e.g. bacteria (Fracchia et al., 2006), winter vomiting disease with *Norovirus* (Myrmel et al., 2006; Van den Berg et al., 2005), *Rotavirus* (Myrmel et al., 2006), hepatitis A virus in Tunisia (Gharbi-Khelifi et al., 2006) –. In tropical climates, human enteric viruses, especially enteroviruses, are isolated throughout the year and in some cases are more prevalent during rainy seasons (Fong and Lipp, 2005). And in Texas, seasonal levels of human enteric viruses in wastewater were highest during late summer, coinciding with the time of substantial crop irrigation: polioviruses were the predominant enteroviruses recovered during spring monitoring, while Coxsackie B viruses comprised the majority of identified isolates recovered in summer (Moore et al., 1988). Similar variations have been obtained for *Campylobacter*, *Salmonella*, vero-cytotoxigenic *Escherichia coli*, *Cryptosporidium* and *Giardia* for various locations in temperate developed countries (Lal et al., 2012). By contrast, *Norovirus* outbreak activity is higher between November and March in the U.S. (Yen et al., 2011). Even with seasonal variations in wastewaters, biofilm may enable the persistence of *Norovirus* and *Enteroviruses* (Skraber et al., 2009). They also depend on **wastewater treatments** (Bixio et al., 2005), agricultural practices (crops, irrigation type (mode, frequency and volumes), local specificities (soil type, wind, tree edges and distance to public paths), and **irrigation method** (Hamilton et al., 2006). They also depend on **the populations** (race, age structure and demography, socioeconomic and educational levels)**, their lifestyles** (food consumption, hygiene and mobility …) (Hamilton et al., 2006), **and their health** that may be taken into account for quantitative microbial risk assessment dealing with the consumption of food irrigated by wastewater (WHO, 2006a). **Sanitary/health hazards also depend on the dilution of wastewater with conventional ones** (Van der Bruggen, 2010).

Biological hazards (viruses, bacteria, protozoa and helminth eggs)

Although not directly related to wastewaters, **waterborne outbreaks caused by enteric pathogens in drinking waters** (Craun et al., 2010; Hoffman et al., 2009; Zhuang et Jin, 2008; Reynolds et al., 2008; Craun et al., 2006; Gerba, 1999) **or recreational waters** (swimming pools, lakes, lagoons, rivers and thermal station) (Sinclair et al., 2009; Hamilton et al., 2007; Craun et al., 2005) **help identify risks of wastewater reuse in irrigation**, since they partly result from the contaminations of conventional water by wastewater. Although drinking water in the U.S. is among the safest in the world, 780 waterborne outbreaks were reported from 1971 to 2006 that were caused by chemical contaminants (11.5%), viruses (8.2%), bacteria (16.6%), protozoa (18.3%), mixed (0.8%) and unidentified agents (44.6%) (Craun et al., 2010). Waterborne outbreaks caused by unidentified agents are mainly acute gastrointestinal illness, the characteristics of these outbreaks being often consistent with a viral aetiology (Reynolds et al., 2008). Outbreaks reported for 1971-2006 have resulted in 577094 cases of illness and 93 deaths (Craun et al., 2010), but the true impact of drinking water contaminations is much higher: Morris and Levein (1995) estimated that **7 million people become hill and more than 1000 die each year in the U.S. as a result of waterborne microbial infection**, and Colford et al. (2006) estimated there to be 4.3-11.7 million cases of acute gastrointestinal illness attributable to public drinking water systems in the United States each year. To our knowledge, there is no equivalent review to those of Craun et al. (2010) and Reynolds et al. (2008) for European countries; partial informations are accessible for countries – e.g. for France (Bonnin et al., 2012; ANOFEL Cryptosporidium National Network, 2010; INVS, 2004; INVS, 2001), for The Netherlands (Svraka et al., 2007) –.

The primary concern in agricultural food consumption deals with uncooked vegetables (Bos et al., 2010; WHO, 2006), e.g. salads, carrots and onions, and restricted irrigation is necessary depending on water quality and management, but it may be hard to implement (Keraita et al., 2010). Other concerns deal with the risk of contamination of farmers or neighbouring inhabitants by bioaerosols emitted during sprinkler irrigation and agricultural work, particularly in windy conditions, and with the risk for human pathogen to reach underground water or surface water bodies without being inactivated (George et al., 2004). Unfortunately, only a few studies have dealt with human contamination though bioaerosol emitted during either sprinkling irrigation, during wind events after sludge application (Baertsch et al., 2007) or flood irrigation (Paez-Rubio et al., 2005), and during activated sludge treatments (Heinonen-Tanski et al., 2009; Fracchia et al., 2006; Sigari et al., 2006) and lead to regulations that are not scientifically sound (Lazarova and Brissaud, 2007); in addition for enteric pathogens transmitted by air way, airborne disease, except for *Legionella pneumophila* and SARS virus, there is a lack of knowledge to assess the actual transfer from the respiratory to the gastro-enteric track and assumptions are performed in quantitative microbial risk assessment on soil particle ingestion and their level of contamination (Mara et al., 2007).

Major human enteric and water-based pathogens

Currently, **more than 140 human enteric pathogens may be transmitted by water route** (Reynolds et al., 2008); they include viruses (Carducci et al., 2009), bacteria (Pachepsky et al, 2011), protozoa (Mota et al., 2009; Tzipori and Widmer, 2008), cyanobacteria (Sivonen and Jones, 1998), and helminth eggs (Gupta et al., 2009; Ensink and van der Hoek, 2006).

Table 1: Most common enteric human pathogens and water-based pathogens in drinking water (The name of water-based pathogens are bold and underlined).

Virus*	Bacteria*	Protozoa*	Helminth eggs**
Norovirus *Sapprovirus* *Paraechovirus* *Enterovirus:* (69-71, *Poliovirus*, *Coxsackievirus*, *Echovirus*) *Reovirus* *Adenovirus* Hepatitis A Virus Hepatitis E Virus *Rotavirus* *Astrovirus* *Picobirnavirus* *Coronavirus*	*Vibrio cholerae* *Salmonella* spp. *Shigella* spp. Toxigenic *Escherichia coli* *Campylobacter* spp. *Yersinia enterocolitica* *Plesiomonas shigelloides* ***<u>Legionella</u>*** *Heliobacter pylori*	*Giardia Lamblia* *Cryptosporidium parvum* *Entamoeba histolitica* *Cyclospora cayetanensis* *Isospora belli* *Microsporidia* *Ballentidium coli* *Toxoplasma gondii* ***<u>Naegleria fowleri</u>***	*Ascaris,* *hookworms,* *Taenia spp.* *Schistosoma* spp.

*: based on Reynolds et al. (2008) for drinking water in U.S. context; **: based on Bos et al. (2010)

Among them, **one has to distinguish** on the one hand **obligatory pathogens** (all human enteric viruses and helminths, several bacteria and protozoa), **from facultative pathogens that may also develop in soil and/or water** – e.g. *Legionella pneumophila* (Berthelot et al., 2009) and Naegleria fowleri (Reynolds et al., 2008) –, on the other hand **pathogens specific to humans from zoonotic pathogens** (Gerba and Smith, 2005). A list of the most common enteric and water-based pathogens in drinking water is proposed in **Table 1**. The same pathogens are also found in food-related illness and death (Mead et al., 1999).

Nearly at the same time in 2008-2009, the U.S. Environmental Protection Agency (US-EPA) and the American Water Works Association (AWWA) each proposed unregulated Contaminant Candidate Lists (CCL) for drinking water by using quite different methodologies according to the same 3 following criteria (Hoffman et al., 2009): (i) the contaminant may have an adverse effect on the human health; (2) it is known to occur or there is a substantial likelihood that it will occur in public water systems with a frequency and at levels of public health concern; and (3) regulation of such a contaminant would present a meaningful opportunity for the reduction of health risk. The final Contaminant Candidate List of the U.S. Environmental Protection Agency in 2009 (U.S.-EPA, 2009) is similar to the List of the American Water Works Association (**Table 2**) with common pathogens: 3 groups of viruses (*Caliciviruses* (includes *Norovirus*) *Enteroviruses* (*Coxsackieviruses* and *Echoviruses*), Hepatitis A virus), 5 groups of bacteria (*Campylobacter jejuni*, *Escherichia coli* (0157), *Legionella pneumophila*, *Mycobacterium avium*, *Salmonella enterica*), and 1 group of protozoa (*Shigella sonnei*). The U.S. Environmental Protection Agency considers *Cyanobacteria* (blue-green algae), other freshwater algae, and their toxins only through toxins in the final Contaminant Candidate List 3 (US EPA, 2009), since they affect human health only through toxin excreted in waters (Sivonen and Jones, 1998).

Table 2: Final Third Drinking Water Contaminant Candidate List (CCL 3) (US-EPA, 2009) and alternative proposal of the American Water Works Association (Hoffman et al., 2009)

Final CCL 3(US-EPA, 2009)	AWWA list (Hoffman et al., 2009)
Adenovirus	
Caliciviruses (includes Norovirus)	*Caliciviruses* (*Norovirus*)
Campylobacter jejuni	*Campylobacter*-like organisms
Enterovirusess (include *Poliovirus*, *Coxsackievirus* and *Echovirus*)	*Enteroviruses* (include *Coxsackievirus* and *Echovirus*)
Escherichia coli (0157)	Toxigenic *Escherichia coli*
Helicobacter pylori	
Hepatitis A virus	Hepatitis A virus
Legionella pneumophila	*Legionella pneumophilia*
Mycobacterium avium	*Mycobacterium avium*
Naegleria fowleri	
	Rotavirus
Salmonella enterica	*Salmonella enterica*
Shigella sonnei	*Shigella spp.*
	Vibrio cholerae

Viruses are obligate, intracellular parasites (Fong and Lipp, 2005; Sobsey and Meschke, 2003). **Most of human enteric viruses** are naked viruses (Fong and Lipp, 2005) that **may survive a long time in water environments** (weeks to months) (La Rosa et al., 2012), with the well-known exception of the severe acute respiratory syndrome (SARS) virus which is an enveloped virus that may be considered as enteric and that is more rapidly inactivated in water and wastewater at ambient temperatures (Gundy et al., 2009). **Most of them are human specific**, except hepatitis E virus with infections suggested from pigs from almost identical viruses (Koopmans and Duizer, 2004) and infection proven from deer (Tei et al., 2003), and the severe acute respiratory syndrome virus which moves from the bat population to other animals and humans (Gundy et al., 2009; Bennett, 2006). Viruses of greatest concern in water (and their associative illnesses) include the group of ***Enteroviruses*** with *Poliovirus*, *Coxsackievirus* and *Echovirus* (diarrhoea, meningitis, myocarditis, fever, respiratory disease, nervous system disorders, birth defects), **hepatitis A virus** (hepatitis, liver damage and jaundice), ***Norovirus*** (diarrhoea), ***Astrovirus*** (diarrhoea), ***Adenovirus*** (diarrhoea, respiratory disease, eye infections. heart disease), and ***Rotavirus*** (diarrhoea) (Rodríguez-Lázaro et al., 2012). In practice, however, the most commonly reported foodborne viral infections are viral gastroenteritis and less frequently hepatitis A (Seymour and Appleton, 2001). Viruses have the greatest infectivity, requiring the fewest number to cause infection of all waterborne microorganisms (Morin and Picoche, 2008), and they are excreted in the faeces in the largest numbers up to 10^{11}-10^{12}.g^{-1} faeces for sick and healthy carriers (Maunula et al., 2013; La Rosa et al., 2012; Da Silva et al., 2010; Reynolds et al., 2008; Fong and Lipp, 2005; Koopmans and Duizer, 2004) and several weeks after the illness period (Maunula et al., 2013; Da Silva et al., 2007). **They are not efficiently removed by conventional filtration and they are more resistant to disinfectants than bacteria** (CFPTEP, 2010a; CFPTEP, 2010b; Aronino et al., 2009; Petrinca et al., 2009; Da Silva et al., 2008), although some studies shows an high efficiency of treatments with differences in the decrease of GI and GII *Norovirus* groups (Da Silva et al., 2007) or of the nearly complete treatment train in the Upper Occoquan Sewage Authority Water Reclamation Plant that protect the Occoquan Reservoir (Rose et al., 2001). Viruses are detected in raw wastewaters as well as in treated wastewaters at the exit of wastewater treatment plants – e.g. in 5 wastewater treatment plants around Rome (La Rosa et al., 2010), in 2 wastewater treatment plants in the southwest of the Netherlands (Van den Berg et al., 2005), for Leipzig (Pusch et al., 2005), for Rio de Janeiro in Brazil (Villar et al., 2007), and in Norway for Oslo area and elsewhere (Myrmel et al., 2006) –. This may explain why they are also found in conventional surface waters and groundwaters. Because of their small size (0.02-0.1 µm) and ease of transport in the subsurface, viruses are of primary concern in groundwaters (Reynolds et al., 2008). They are known to be the causative agent in 8.2% of drinking water outbreaks reported in recent years in the U.S. (Craun et al., 2010) to which probably most of the 44.6% of outbreaks of undetermined etiology have to be added. The overall method to extract and detect viruses in foods using molecular tools could be divided into three different steps: (1) virus elution and clarification from substrates, (2) concentration of the viruses (Hamza et al., 2009; Croci et al., 2008; Liu et al., 2007; Villar et al., 2006; Dubois et al., 2006; Katayama et al., 2002; Jothikumar et al. 1995; Tsai et al., 1993), and (3) RNA or DNA extraction and purification, amplification, detection of amplified products, and confirmation of the results (Croci et al., 2008). For each of these steps various methods exist. Among the methodological difficulties: the easiness to obtain GC enumerations but the difficulties to estimate the proportion of viruses that remain infectious (Nuanualsuwan and Cliver, 2002), although some PCR methods are proposed to discriminate between infectious and non-infectious viruses (Bhattacharya et al., 2004; Nuanualsuwan and Cliver, 2002).Other detection methods exist, including virus cultivation that enables to enumerate infectious viruses, but that is also fastidious and that is still not possible for several viruses including *Norovirus*. Due to the potential great concern of viruses in raw and treated wastewaters, we add to this review a more complete in **Appendix 1**.

Bacteria are prokaryotic, single-celled organisms, ranging in size from 0.1 to 10 µpm (Reynolds et al., 2008). Enteric bacteria are able to colonize the human intestinal and gastrointestinal tract. Generally, **enteric bacteria do not survive long in the environment, although some have resistant spores or can form dormant stages** that aid in their survival. Waterborne outbreaks caused by enteric bacteria primarily occur because of failed or absent treatment processes. Waterborne enteric bacteria of greatest concern in water (and their associative illnesses) include *Salmonella* (typhoid, diarrhoea), *Shigella* (diarrhoea), *Campylobacter* (diarrhoea, nervous system disorders), *Vibrio cholerae* (diarrhoea), and enterotoxigenic *Escherichia coli* (*via* produced toxins: diarrhoea, kidney failure, haemorrhagic colitis) (Gupta et al., 2008), *Helicobacter pylori* (duodenal and gastric ulcers, infections can lead to gastric cancer (Reynolds et al., 2008)), and *Mycobacterium avium* (lung infection, disseminated infection in severely immunocompromised peoples). Although the contribution of the waterborne route of exposure to the disease is uncertain, studies have found 10%-60% of individual groundwater wells contaminated with *Helicobacter pylori* (Park et al., 2001). *Legionella* pneumophila (lung diseases) is an important water-based bacteria that may develop in stagnant water at warm temperature (Reynolds et al., 2008); in the U.S., 6 water-associated outbreaks were recorded in 2001-2002 (Reynolds et al., 2008). Other water-based pathogens include *Leptospira* spp. (Leptospirosis) and *Burkholderia pseudomallei* (acute and chronic forms of melioidosis; symptoms may include pain in chest, bones, or joints; cough; skin infections, lung nodules and pneumonia) (endemic areas include particularly Thailand and northern Australia).

Enteric protozoa are single-celled animals that live in the gastrointestinal tract of infected individuals (Reynolds et al., 2008). They range in size from 1 to I00 µm and produce an environmentally stable cysts or oocysts that survive for long intervals in wastewater, aquatic and terrestrial environments. The thick cyst or oocyst walls are highly resistant to disinfectants used in conventional water treatment. Waterborne enteric protozoa of primary concern in water (and their associative illnesses) include *Cryptosporidium* (*parvum*, *hominis* …) (Xiao, 2010) and *Giardia* (*lamblia*, *Instestinalis* …) (Amahmid et al., 1999); they are implied in dysentery diseases, infections of the liver, lungs, pericardium, skin and brain. A well-known example of *Cryptosporidium* outbreak is the outbreak that took place in Milwaukee, Wisconsin in 1993 (Hoxie et al., 1997; Mac Kenzie et al., 1994). *Entamoeba histolytica* is another parasite that causes 40000-100000 deaths annually in the World (Ackers and Mirelman, 2006); in the large bowel, infections that are exclusively luminal are asymptomatic, and clinical amoebiasis only occur when the parasite penetrates the colon wall, causing flask-shaped ulcers that lead to amoebic dysentery. Much less frequently, Entamoeba spread through the portal vein to the liver (amoebic liver abscess) and, very rarely, disseminate to other sites. *Cyclospora caryentensis* is another parasite that has been linked to a possible waterborne outbreak in the U.S. (Mansfield and Ciajadhar 2004). *Naegleria fowleri* (primary amoebic meningoencephalitis) is a water-based pathogen of primary concern because of a high fatality rate in diagnosed cases. Two deaths occurred in an outbreak of *Naegleria* in 2002 (Reynolds et al., 2008). Cysts of the three species are resistant to desiccation, temperature, pH variations and chlorination. Enteric protozoa are of major concerns for operators involved in recycling wastewaters; they may be easily removed by filtrations.

While helminths are a major problem in some countries (Gupta et al., 2009; Ensink and van der Hoek, 2006; Amahmid et al., 1999), they seem less and less important in others, to such an extent that helminth eggs have disappeared from the French standards in 2010 (MSS, 2010). The most common species are *Ascaris lumbricoides*, hookworms, *Enterobius vermicularis*, *Trichostrongylus spp.*, *Taenia* spp., *Hymenolepis nana* and *Dicrocoelium dendriticum*.

Quantitative microbial risk assessment

Quantitative microbial risk assessment (QMRA) tools permit *a priori* to assess the risk of exposure, infection and disease of peoples via pathogens in food, water or air (Schijven et al., 2013; Pachepsky et al., 2011; Hunter et al., 2003; Petterson and Ashbolt, 2003). They can take into account the disappearance of pathogens over the entire chain from water treatment to the potential exposure of peoples (Mara, 2011; Mara et al., 2010; Stine et al., 2005). Considering a number of disability adjusted life years (DALY) per case of disease (pcd) that may vary with regions (Institute for Health Metrics and Evaluation. 2013; WHO, 2006a; WHO, 2004), these tools allow to compare the effects of different pathogens, to take into account different pathogens simultaneously, and to achieve tolerable targets of disability adjusted life years (DALY) per person per year (pppy) by satisfying required microbial reductions either exclusively during wastewater treatment or by combining the effects of treatment to pathogen decay after irrigation before and after harvest (WHO, 2006a; Stine et al., 2005).

The quantitative microbial risk assessment is based on the initial mathematical model for drinking water (Fewtrell and Bartram, 2001; Haas et al., 1999; Haas et al., 1993) issued from chemical risk assessment approaches. It combines 3 types of considerations (Haas et al., 1999): the exposure assessment (i.e. the number of pathogen ingested during an exposure event through food, water or air ways), the dose-response assessment (i.e. the relationship between infection probability and the number of pathogens ingested), and risk characterization (i.e. the probability of illness (see death) for infected peoples). A framework to adapt to country or regional contexts has been proposed (Scheierling et al., 2010; WHO, 2006a). Considering a tolerable disability adjusted life years per person per year and the disability adjusted life years per case of disease, one has (i) to estimate the required level of reduction of pathogens initially present in raw wastewater (step 1: calculation of the tolerable disease and infection risks per person per year; step 2: calculation of the tolerable infection risk and dose of ingested pathogens per exposure event; step 3: calculation of the level of reduction of pathogens required to achieve the health targets), (ii) specify how pathogen reduction would be achieved by wastewater treatment commonly in conjunction with other heath protection measures, and (iii) to verify that the targets have been achieved through bioindicators or pathogen themselves. Deterministic models of quantitative microbial risk assessment have been proposed (Hamilton et al., 2007a), but quantitative microbial risk assessment tools may be combined with stochastic simulations (Karavarsamis and Hamilton, 2010; Mara and Sleigh, 2010; Mara et al., 2007; Hamilton et al., 2006; Hamilton and Stagnitti, 2006; Sleigh and Mara, 2003) to account for the random distribution of some of the context parameters, e.g. the structure of the population (age, body mass, socioeconomic, racial, and geographic demography) and the depending daily consumption (US-EPA, 2003; US-EPA, 1997).

The World Health Organization proposed this tool to help countries develop their own regulations and standards (WHO, 2006a), partly in response to criticism of too liberal previous guidelines in 1989 (WHO, 1989) compared to those of the U.S. Environmental Protection Agency (USEPA/USAID 1992). This recommendation was based on some works (Fattal et al., 2004; Sleigh and Mara, 2003; Blumenthal et al., 2003; Blumenthal et al., 2000; Tanaka et al., 1998; Shuval et al., 1997; Asano et al., 1992 among others) and it followed the adoption of QMRA in the Australian national water recycling guidelines (EPHC/NRMMC/AHMC, 2006). Additional applications of quantitative microbial risk assessments to wastewater reuse for irrigation have been published later. They give a first overview of risks resulting from the ingestion of crops irrigated by wastewater as a function of the pathogens and their concentrations in the effluent – e.g. viruses (Mara and Sleigh, 2010c; Mara and Sleigh, 2010b; Mara et al., 2007; Hamilton et al., 2006; Stine et al., 2005; Petterson et al., 2001), bacteria (Stine et al., 2005), protozoa (Mota et al., 2009), and helminths (Mara and Sleigh, 2010a; Mara and Sleigh, 2010b) –, the crops – e.g. lettuce (Mara et al., 2007; Hamilton et al., 2006; Stine et al., 2005; Petterson et al., 2001),cucumber (Hamilton et al., 2006), broccoli (Hamilton et al., 2006), cabbage (Hamilton et al., 2006), cantaloupe (Stine et al., 2005), and pepper (Stine et al., 2005) –, the irrigation type (subsurface, furrow, or drip irrigation), and other practices such as the delay between the last irrigation and the harvest, postharvest washing/disinfection, and the preparation of food (Hamilton et al., 2006). Inhalation (and the involuntary ingestion) by farmers or neighbouring inhabitants of aerosolized soil particles have been discussed for highly mechanised and manual cropping systems (Mara et al., 2007). Some papers proposed updates from the WHO guidelines (Mara et al., 2010), and Maimon et al. (2010) proposed a review on the risks resulting from different current guidelines.

The value for the tolerable DALY (10^{-6} pppy) proposed in the WHO guidelines (WHO, 2006a) and in the Australian national water recycling guidelines (EPHC/NRMMC/AHMC, 2006), as currently retained as for drinking water (WHO, 2006a), may be too restrictive in most of the developing countries (Mara and Sleigh, 2010c), and other threshold values have been proposed: 10^{-5} (Mara and Sleigh, 2010c) and 10^{-4} pppy (Mara, 2013; Mara, 2011; Mara and Sleigh, 2010b; Stine et al., 2005) among others. On the one hand, other routes of contamination can largely predominate – e.g. even with raw wastewater used for spinach and Cauliflower irrigation in Pakistan, unhygienic post-harvest handling was the major source of produce contamination (Ensink and van der Hoek, 2006), and other contaminations in the market may result from the use of contaminated freshening water (WHO, 2006a) –. On the other hand, a too low threshold value would lead to additional costs that may be disproportionate with regard to the expected health gain and unrealistic for several developing countries (Blumenthal et al., 2000). An increase in the tolerable DALY per person per year from 10^{-6} to 10^{-5}-10^{-4}, would then lead to very simple wastewater treatment systems to achieve a single-log unit pathogen reduction as the balance of the required total pathogen reduction (i.e. 10^{3}-10^{5}) can be easily achieved by very reliable post-treatment health-protection control measures (pathogen die-off and produce washing or disinfection) (Mara and Sleigh, 2010b-c). As proposed by the World Health Organization (WHO, 2006), tolerable DALY should be adapted to national or regional contexts, and possibly distinguish agricultural production for local consumption from those for export. The Californian standard of $\leq$2.2 total coliforms (roughly $\leq$1 E. coli) per 100 ml and the recommendation of USEPA & USAID (1992) for an ‘undetectable’ level of E. coli in 100 ml result in *Rotavirus* infection risks of 10^{-6}-10^{-8} pppy. Such low levels of risk are difficult to justify epidemiologically, and they are unlikely to be cost effective in protecting health (Mara et al., 2007). However, the usefulness of quantitative microbial risk assessment is dependent upon the quality and appropriate use of available data for describing the occurrence, persistence and human dose-response of pathogens (Petterson and Ashbolt, 2003). First, QMRA approach requires to correctly measure or estimate the initial amounts of pathogens in the raw wastewater and their actual fates during wastewater treatments, storage in ponds or reservoirs, distribution of reclaimed wastewater and after irrigation (before and after harvest); processes are still misunderstood and/or poorly quantified (virus internalization in edible parts of crops eaten crude (Wei et al., 2011; Urbanucci et al., 2009; Chancellor et al., 2006; Oron et al., 1995) and virus attachment at the surface of vegetables (Vega et al., 2005); pathogen transfer from respiratory track to gastro-enteric track among others as implicitly assumed by Mara et al. (2007) and demonstrated in other contexts (Marks et al., 2000), as pathogens may be present in bioaerosols (Fracchia et al., 2006; Carducci et al., 2000; Brandi et al., 2000); the effect of washing edible parts of foods (Mara, 2013; WHO, 2006a among others) whose effects may be overestimated with regard to fine works on works on some crops (Gerba and Kennedy, 2007) and on other types of surfaces (Barker et al., 2004)), and several constants remain imprecise as those to estimate pathogen decays during and after treatment (Mara, 2013). Second, estimating exposure, infection and disease risks simultaneously requires having a good description of human comportments (nutrition, sanitary habits, movements and body protection …) as partly available for some countries (U.S. Environmental Protection Agency, 2003; U.S. Environmental Protection Agency, 1997) but not everywhere. Third, real time detections of actual human pathogens remains difficult and expensive, although great progresses have been done in molecular methods, and the enumeration of bioindicators instead of actual pathogens may be criticized (see below); this last point is complex to solve as 'reference' pathogens (e.g., *Norovirus*, *Campylobacter*, *Cryptosporidium*, Ascaris) can be chosen but tolerable DALY pppy may led to pathogen concentrations so low especially for viruses – e.g. $5\ 10^{-3}$ Rotavirus/L (WHO, 2006a) – that they cannot be detected without preliminary great concentrations. Bioindicator concentrations are usually greatly higher but their correlation with actual pathogen concentrations and fates is questionable (see below).

Similarly as for microbial risks, quantitative chemical risk assessment may be applied to organic pollutant in wastewaters. The approach may be based on acceptable concentration of pollutant, i.e. zero effect concentration, rather than disability adjusted life years (DALY) (Weber et al., 2006). And other approaches of risk assessment combining physiochemical, environmental properties and toxicity of the 200 most commonly prescribed drugs, as well as common antibiotics and lipid regulating drugs that were not within the top 200, has been used to rank different risks (Cooper et al., 2008). Unfortunately and similarly to microbial risk assessment, there is currently a lack of information concerning the effects of antibiotics to critically assess potential risks for environmental discharge and water recycling (Watkinson et al., 2007).

Bioindicators of human pathogens

Bioindicators are used as indexes of the presence and concentration of human pathogens in raw and treated wastewaters as well as in conventional, recreational and drinking waters; **they are also used to assess/control the reliability of plant treatments and identify the origin of water**. Their use can be justified by the great number and the diversity of human pathogens in waters, their dangerousness, and difficulties in their enumeration; **bioindicators are easier, faster and cheaper to isolate, cultivate and identify** (Keegan et al., 2009). Moreover, **pathogen concentrations not-measurable without first concentrate the**

suspensions are often beyond maximum tolerable thresholds that may be calculated by quantitative microbial risk assessment (QMRA) for a tolerated disability adjusted life years (DALY) of 10^{-6} per person per year; this is especially the case for human enteric viruses – e.g. 5 10^{-3} *Rotavirus*.L^{-1} proposed in an example proposed by the World Health Organization (WHO, 2006a) –. By contrast bioindicators are more abundant; the same example assumes that there are 10^5-10^7 *Escherichia coli* per *Rotavirus* (WHO, 2006a). The most common indicators for microbial water quality and human health risk assessment are total coliform, faecal coliform or *Escherichia* coli, and faecal enterococcus (Levantesi et al., 2010; Ministère de la Santé et des Sports, 2010; Jiang, 2006); other ones include spores of anaerobic sulphite-reducing bacteria (Ministère de la Santé et des Sports. 2010), *Clostridium* spores (Levantesi et al., 2010) and F-specific RNA phages (Ministère de la Santé et des Sports. 2010). To address some of their deficiencies outlined below, others bioindicators have been proposed without being retained to our knowledge: somatic coliphages and *Bacteroides* phages to replace the F-specific phages (Leclerc et al. 2000), *Escherichia faecalis* to replace *Escherichia coli* (Fujioka and Yoneyama, 2002). **Unfortunately, no bioindicator can mimic the behaviour and characteristics of the true pathogens** (Payment and Locas, 2011; Symonds et al., 2009; Keegan et al., 2009; Petrinca et al., 2009; Salgot et al., 2003; Sinton et al., 2002; Moore et al., 1988), only about 3% of individual humans carry the F-specific RNA phages and their abundance in wastewater treatment plants may result from their multiplication in sewage (Leclerc et al. 2000), and there is little concordance in the sample volumes (from 1 to 400 L for bacteriophages), in the concentration methods and in the phage detection methods, thus making comparisons extremely difficult (Leclerc et al. 2000). Several works noted no or poor relationships between classical faecal bioindicators and pathogens in drinking waters (Figueras and Borrego, 2010), in sewage and/or reclaimed wastewaters (Payment and Locas, 2011; Levantesi et al., 2010; Carducci et al., 2009; Haack et al., 2009; Villar et al., 2007; Harwood et al., 2005; Leclerc et al. 2000), in surface waters (Payment and Locas, 2011; Skraber et al., 2004; Metcalf et al., 1995), in groundwater (Payment and Locas, 2011;), and in treated wastewater for irrigation (Holvoet et al., 2014). More precisely **in sewage, even with very high levels of microorganisms, no mathematical correlation can predict the type or concentration of any pathogen** (Payment and Locas, 2011). Difference between pathogen and bioindicator resistances to plant treatments contribute also to the lack of correlation – e.g. *Adenoviruses* have been found to be significantly more stable than faecal indicator bacteria and other enteric viruses during UV treatment (Jiang, 2006); *Escherichia coli* is so sensitive to sunlight inactivation that it should not be used to monitor recreational waters for the presence of human enteric viruses (Fujioka and Yoneyama, 2002) –. **After discharge in the environment, direct correlation becomes biologically improbable** as dilution, transport, and different inactivation rates occur in various environments (Payment and Locas, 2011). **This may lead to false alert and to lack of alert,** as at Milwaukee where drinking water contamination lead over 400000 people to be infected with cryptosporidium in 1993 (Mac Kenzie et al., 1994). However, bioindicators are still useful as a measure of risk (Holvoet et al., 2014; Payment and Locas, 2011). Moreover, several authors noticed that bioindicators may be useful **to assess/control the reliability of plant treatments** while mentioning differences between the fates of bioindicators and pathogens during some treatments (Lucena et al., 2004; Vilanova et al., 2004; Ashbolt et al., 2001). Today, we can assume that the monitoring of bioindicators may be replaced shortly by the direct detection of pathogenic microorganisms (Figueras and Borrego, 2010). , including for example human enteric viruses (Bosch et al., 2008). New developments in molecular techniques with real-time or quantitative PCR, Multiplex PCR and genetic microarrays would probably enable faster determination of pathogen themselves, with much more information on their viability and infectivity (Yeh et al., 2009; Stratton and Matthews, 2009). Especially, methods for virus extraction, concentration, and enumeration have quickly evolved during the last 10 years (Mattison et Bidawid, 2009; Rodríguez et al., 2009b; Hamza et al., 2009; Bosch et al., 2008; Croci et al., 2008; Morin and Picoche, 2008; Oillic et al., 2007; Da Silva et al., 2007; Liu et al., 2007; Villar et al., 2006; Dubois et al., 2006; Villar et al., 2006; Dubois et al., 2006; Fong et Lipp. 2005; He et Jiang, 2005; Katayama et al., 2002; Jothikumar et al. 1995; Tsai et al., 1993).

Chemical hazards

Raw wastewaters may contain various chemicals including mineral compounds (N compounds, phosphates, chloride, sodium, borate, trace metals …) (Leverenz et al., 2011; Toze, 2006b; Unkovich et al., 2006), and organic pollutants (Toze, 2006b; Toze, 2006a). N-compounds and phosphates may favour river eutrophication (Thomas et al., 2010; WHO-EC, 2002), while they are important nutrients for crops. Possible **high contents in sodium cation have detrimental effects on the structural stability of soils**, their compactness and their water permeability (Mazzini et al., 2013), and **high concentrations in sodium, chloride and borate ions are toxic for plants** (Unkovich et al., 2006). Heavy metals are of little concern for irrigation with treated wastewaters as most of those in raw sewage are immobilized on the biosolid fraction (sludge …) during classical treatments (Toze, 2006b). However, some authors recommended to use an additional treatment process to decrease the level of metals in the sewage irrigation water (Afifi et al., 2011) and, when wastewater is of industrial origin and/or is not sufficiently treated, heavy metals would need to be considered (Toze, 2006b; Foster and Chilton, 2004), notably lead, chromium, cadmium (Foster and Chilton, 2004), – e.g. as in India (Sridhara Chary et al., 2008) and probably in the past in China, with as a lasting effect due to the very low mobility of metals in soils (Khan et al., 2008) –. Organic pollutants are often considered as 'emerging pollutants', i.e. new chemicals without regulatory status and which impacts on environment and human health are poorly understood for the U.S. Environmental Protection Agency (Deblonde et al., 2011). **Organic pollutants from house wastewaters include chemicals of personal care products, various pharmaceutical products** (antibiotics, lipid regulator agents, anti-inflammatory drugs, β-blockers, cancer therapeutics, contraceptives and other hormones) (Barnes et al., 2008; Chefetz et al., 2008; Al-Rifai et al., 2007; Nikolaou et al., 2007; Conn et al., 2006; Hernando et al., 2006; Karthikeyan and Meyer, 2006; Barnes et al., 2004)**, bisphenol A and phthalates** (Barnabé et al., 2008; Clara et al., 2010; Dargnat et al., 2009). When a unique urban sewer network collects both house wastewaters and runoff waters during rainy events, w**astewaters may also contain pesticides** (Gasperi et al., 2008b; Blanchoud et al., 2007) **and polycyclic aromatic hydrocarbons** (Palmquist and Hanaeus, 2005). In addition, **solvents are produced during disinfection by chlorine** (Kim et al., 2003). And **neurotoxins and carcinogenic hepatotoxins may be synthesized by cyanobacteria** (Sivonen and Jones, 1998) from which blooms are favoured by wastewater (Barrington and Chadouani, 2008); in several countries, water coming from ponds without special treatment contains microcystins that have an impact both on the growth and the development of crops (Gehringer et al., 2003; Saqrane et al., 2008). C**oncentrations of most organic pollutants in urban wastewaters** (raw or treated) **are generally below the toxic level**s for humans, but potential **problems may result from the combined effects of several pollutants and/or their cumulative consequences over long-term periods**, all the more that some pharmaceuticals and personal care products are very persistent in the environment and/or can accumulate in plants. The great diversity of compounds leads to fragmentary knowledge on their environmental fate and impacts. Impacts on aquatic ecosystems have been the most discussed (Gros et al., 2010). Impacts on human health remain uncertain for irrigation reuse. They may *a priori* result from pharmaceutically active compounds and a large number of compounds that are known or suspected endocrine disruptors: they include the estradiol compounds commonly found in the contraceptive pill, phytoestrogens, pesticides, industrial chemicals such as bisphenol A and nonylphenol, and heavy metals (Toze, 2006b). Groundwater contamination may be direct from residential septic systems (Swartz et al., 2006). The removal efficiency of treatment trains in wastewater treatment plants varies between compounds and treatments trains (Watkinson et al., 2007), and several studies noted the presence of organic contaminants in the environment – e.g. antibiotic in watershed (Watkinson et al., 2009) –. Due to the potential concern of organic pollutants in raw and treated wastewaters, we add to this review a more detailed review on the diversity and the environmental fate of organic contaminants in **Appendix 2**.

Environmental and agricultural hazards

Irrigation with wastewater partly protects rivers, aquifers and the sea from direct discharge of wastewaters into these water bodies. Other well-known environmental benefits include the recovery of nutrients for agriculture, and the sustainability of water resource management.

However, **the use of untreated wastewater has led sometimes to local or regional disastrous effects**. As a European example, Milan is the largest European project for agricultural irrigation reuse. Irrigation is taken from streams and a network of intermeshed ditches and canals developed by Cistercian monks in which raw wastewaters were discharged until recently; the system has been completed with derivation canals from Lugano and Maggiore lakes. The doubling of Milan population since the late 19th century together with 1 M daily workers has led to huge amount of wastewater, loss of permeability and contamination of soils, and 30% of the pollution in the northern portion of the Adriatic Sea south of Venice, until Nosedo and San Rocco wastewater treatment plants have been operational in 2003 and 2004, respectively (Mazzini et al., 2013).

Environmental hazards may result from the salinity and sodicity of wastewaters that affect soil structural stability, soil structure and soil mechanical strength (Sou/Dakouré et al., 2013; Qadir and Drechsel, 2010; Vogeler, 2009; Tal, 2008; Gharaibeh et al., 2007; Toze, 2006; Gerba and Smith, 2005) and indirectly soil air and water permeabilities, as well as soil aeration. The excessive salinity and sodicity of wastewaters lead sometimes farmer associations to mix water desalinized by reverse osmosis to other treated wastewater to insure acceptable water electrical conductivity (Renault et al., 2013). Hazards also result from the toxicity for plants of high salinity and high levels of chlorine, sodium and boron (Tal, 2008; Hamilton et al., 2007; Unkovich et al., 2006; GWRC Report, 2005; Angelakis et al., 1999). At least wastewaters may contain toxic compounds, by emerging organic substances (disruptors endocrine, pharmaceuticals including antibiotics ...) (Leverenz et al., 2011, Barnes et al., 2008, Al-Rifai et al., 2007; Karthikeyan and Meyer, 2006). They may affect the environmental biodiversity (), the growth and yield of crops, and/or accumulate in plants (Dolliver et al., 2007; Boxall et al., 2006; Kumar et al., 2005) or animals (Rimkus et al., 1997).

Indirectly, wastewater reuse may induce several changes that have to be explicitly taken into account for decision with cost-benefit approaches: drinking water supply, recreational activities, jobs ….

Economic sustainability of wastewater reuse in irrigation: an overview

Cost-benefit analyses are important to insure the economic sustainability of wastewater reuse for crop irrigation **and optimize the choice of expenditure items**. As an example; the city of San Diego is now going to reuse indirectly wastewater as potable water (Steirer and Thorsen, 2013), since costs of potable reuse are lower than costs of nonpotable reuse and desalination, and cost of importing water which remains smaller is anticipated to rise faster and to become comparable to reuse by 2030 (Trussell et al., 2012). Other costs-benefits analyses have been performed at regional or country level – e.g. in Israel (Haruvy, 1997) –. On the one hand for human health protection, **it is important to know whether additional expenditures must be made, and where they must be made** (on wastewater treatments, on the production of agricultural products and their commercialization, at the level of consumers). It is important to take into account simultaneously all positive or negative impacts on crops (e.g. their yields and qualities), on the environment (e.g. the biodiversity in water bodies, the structural stabilities of soils and their effects on soil compaction, permeability to water, aeration ...), and indirectly on other sectors of the economy (tourism, recreational activities, employment …). On the other hand, **it is important to know how to use economic incentives to promote the reuse of wastewater**, including tariffs, subsidies and taxes (Van der Bruggen, 2010). Although the 'polluter pays' principle is widely accepted in most OECD countries, polluters in Europe currently pay only for treatments required for the discharge of wastewater in conventional water bodies; the cost of additional (regenerating) treatments required to remove remaining contaminants before reuse can be charged partly or totally to farmers or public authorities. It may increase the economic burden of wastewater reuse in agriculture beyond the point which most agriculture crops can carry and it may result in blocking the rational economic development of water recycling and reuse. Therefore in several places, regenerated water is sold at very low prices, e.g. in Milan (Mazzini et al., 2013), Cyprus (Papaiacovou et al., 2013) and Tunisia (Qadir et al., 2010b).

Economic considerations begin to participate to decisions by comparing cost of additional treatments (Molinos-Senante et al., 2013; Iglesias et al., 2010) to direct and indirect benefits (Lavee, 2013, 2011; Molinos-Senante et al., 2010). Such analyses can be performed at national or regional levels by **comparing alternative scenarios** (Lavee, 2013). **Costs of additional water treatments may be estimated** (Molinos-Senante et al., 2013; Iglesias et al., 2010) in order to optimize treatment technologies. Operational costs deal with energy, chemicals, labour, maintenance and sludge and grit disposal (Mazzini et al., 2013). Total cost per m^3 of treated wastewater is usually much higher than the plant unit is small (Hernández-Sancho and Sala-Garrido, 2009), but it greatly depends on the local context, making comparisons difficult between wastewater treatment plants. France has many very small wastewater treatment plants (Golla et al., 2010): in 2011, only 17.3 % of the 19300 plants corresponded to French agglomerations of more than 2000 equivalent habitants, while 17% of the stations deal with more than 90% of the pollution produced (MEDDE, 2013). Beyond about 5.4 million homes were not connected to the public sewage in 2012 (MEDDE, 2012). Decision support systems (DSS) with monetary valuations of costs exist for the selection and design of wastewater treatments (Hamouda et al., 2009; Hidalgo et al. 2007), but there is still a need to develop integrated decision support systems that are generic, usable and consider a system analysis approach (Hamouda et al., 2009).

Taking into account the impact of practices on health, the environment and crops requires to assign a monetary value to these effects. Monetary valuation of disease gave place to several papers about 20 years (Remoundou and Koundouri, 2009). By contrast until recently, monetary valuation of environmental benefits were restricted to some items – e.g. nitrate leaching (Haruvy, 1997)) –, whereas many social and environmental costs have been difficult to quantify (Hamouda et al., 2009). Since a few years, new methods of monetary valuation have been proposed, including contingent valuations (Dupont, 2013; Alcon et al., 2012), estimation of shadow prices for the pollutants removed in a treatment process (Hernández-Sancho et al., 2010; Molinos-Senante et al., 2010) and others (Lavee, 2011).

The use of such methods leads already to some interesting results although their generic value is questionable. As a first example, Shuval (2008) combined a quantitative microbial risk assessment to a cost effectiveness analysis: he estimated that treating wastewater to meet the US-EPA/US-AID guidelines (US-EPA/US-AID, 1992) would result in an additional cost, of some $ 500 000 to $ 1 000 000 per case of disease prevented compared to WHO standard. As a second example, Lavee (2011) examined the costs and benefits associated with possible alternative wastewater treatment standards in Israel; it was found that switching from the current standards to more demanding standards would indeed achieve a net benefit to the national economy, estimated at US\$0.1235/m^3 of treated wastewater. Of course, the results of Lavee greatly differ from those of Fine et al., (2006) who didn't account for environmental impacts.

Therefore, **it seems important to develop integrated decision support systems that are generic, usable and consider a system analysis approach** (Hamouda et al., 2009). And in their objectives for updating their guidelines, the U.S. Environmental Protection Agency would like to have focus on international project economic benefits (Bastian, 2012).

Regulations: the need for stronger scientific basis

Some 50 countries in the world irrigate about 10 million hectares of crops with raw/untreated wastewater producing about 12% of the world food crops (Shuval, 2008); they include China, Mexico, India, Chile, Syria, Pakistan, Colombia, Argentina, several USA states, Ghana, Vietnam, Peru, Turkey, Morocco, Egypt, Kuwait, Sudan, Tunisia, Nepal and Bolivia, although several of them also use treated wastewater in irrigation (Keraita et al., 2008). China, India, Pakistan and Mexico are among the largest countries in this group, and also those most often mentioned for large-scale industrial water pollution and irrigation with highly polluted water (Jiménez and Asano, 2008). **This practice is made possible by the lack of guidelines, regulations, and standards** or their non-compliance; it provides vital work for hundreds of thousands of poor farmers and essential food for malnourished populations, but it often results simultaneously in massive disease transmission (Shuval, 2008). **In Europe, several countries have no regulations or guidelines** dealing with reuse for irrigation: Austria, Czech Republic, Denmark, Estonia, Finland, Iceland, Ireland, Latvia, Lithuania, Luxembourg, Norway, Slovenia, Slovakia, Sweden, Switzerland, and the Netherlands. We can add to this list several others that are still contemplating regulations or guidelines (Belgium, Bulgaria, Hungary, Malta, Poland, Romania, and UK) (Angelakis, 2012), Malta criteria being already in preparation since 2011 (Angelakis, 2012). The lack of regulations/guidelines in these countries generally results from the recycling of wastewater for other uses – e.g. in industry in Belgium (Bixio et al., 2008) – or from the lack of reuse. A probable exception is Bulgaria that has still no regulation, while its water stress index is higher than 60% (Bixio et al., 2005), but there have not been any specific investigations related to wastewater reuse in this country (Angelakis et al., 2007). By contrast, **other European countries have regulations or guidelines dealing with wastewater reuse in irrigation: Italy** (since 1976 with the water Protection Act in 1976 that was replaced in 2003 by the Ministry Decree, D.M. n°185/03 (Mazzini et al. (2013))), **France** (since 1991 (CSHPF, 1991) modified with an 'arrêté' in 2010 (Ministère de la Santé et des Sports, 2010) and currently discussed to be improved again), **Germany** (since 1999 (Jiménez and Asano, 2008)), **Cyprus** (since 2005 (Decree 269/2005) for small agglomerations completed in 2007 with standards set through disposal permits for large agglomerations (Decree 263/2007)), **Portugal** (since 2006, standards NP 4434 (Marecos do Monte, 2010; Marecos do Monte, 2008)), **Spain** (since 2007) (Angelakis, 2012), and **Greece** (since 2011 (Papaiakovou et al., 2011)).

Some reviews have traced the history of the regulations on wastewater reuse in irrigation (Angelakis, 2012; Paranychianakis et al., 2011; Papaiakovou et al., 2011; Shuval, 2008), and the recent guidelines of the U.S. Environmental Protection Agency give an insight into regulations or guidelines and standards of different countries/states (US-EPA, 2012). **Wastewater reuse for irrigation was practiced in the World without regulations or guidelines before 1918**. At this time, the California State Board of Public Health promulgated the initial *Regulation Governing Use of Sewage for Irrigation Purpose*, pertaining to irrigation of non-edible and cooked crops with sewage effluents (Angelakis, 2012; Asano, 2006; Ongerth and Jopling, 1977) and, in 1933, the State Department of Health in California allowed the irrigation of vegetables if the wastewater was oxidized (made non-putrescible) and reliably disinfected or filtered to meet bacterial standard is approximately the same as the current drinking water standard. California has continually revised its regulations and standards since that time to address additional applications, advances in treatment technology, and increased knowledge in microbiology and public health protection (Angelakis, 2012; Crook and Surampalli, 1996; Ongerth and Ongerth, 1982), and publish the so-called Title 22 (State of California, 2000) and the Purple Book (State of California, 2001), which are **a collection of guidelines, rules, and standards corresponding to a "zero tolerance"** (e.g. 2.2 TC/100 mL of treated wastewater for unrestricted irrigation of vegetable crops normally eaten raw). They have been used later elsewhere as basis for regulations and standards (Bixio et al., 2008). Currently, California has the most comprehensive regulations pertaining to the public health aspects of reuse (Zhang, 2012). In recognition of the value of reclaimed water, the U.S. Environmental Protection Agency published guidelines for wastewater reuse, with progressively more stringent standards (initially proposed in 1980, then updated in 1992, 2004 and 2012 (US-EPA, 2012; Crook and Surampalli, 1996). Standards proposed in 1992 together with the US Agency for International Development for unrestricted irrigation include 'No FC detection in 100 mL' (US-EPA/US-AID, 1992), regardless of technical feasibility, of cost effectiveness for other areas of the world, and of the "natural" river water or water at approved bathing beaches in the United States or Europe microbial quality. The State of Florida already adopted 'No detectable E. coli/100 ml for crops consumed raw' in 2003 (Papaiakovou et al., 2011). In the U.S., 25 states had regulations, 16 guidelines, 9 nothing governing the practice of reuse of treated wastewater covering several but not all uses of wastewater in 2003 (Bastian, 2012). **Some other countries have adopted a "zero risk" regulations**, including Israel with its first regulations in 1952 (Shuval, 2008; Brissaud, 2008; Asano, 2006) and increasing with its revision in 1978 and 1999 (i.e. <1 FC/100 ml for unrestricted irrigation in 1999), in Greece (Papaiakovou et al., 2011; Hochstrat et al., 2011) and Australia before 2006 (ARMCANZ-ANZECC-NHMRC, 2000); **such option is generally not economically or technically feasible in developing countries** (Shuval, 2008). In Europe, Italy and Cyprus have established the highest standards, but not as severe as U.S. standards. Cyprus imposes < 5 faecal coliforms/100 mL in 80% of samples for unrestricted irrigation for small agglomerations. In Italy, it is interesting to note the Law decree n°152 in 2003 (Ministry Decree, D.M. n°185/03) set less restrictive rules (<10 E. coli/100 mL in 80% of samples for unrestricted irrigation than the water Protection Act in 1976 (<2 total coliforms in all samples before for unrestricted irrigation). **By contrast, the World Health Organisation (WHO) proposed first in 1973 Guidelines** with standards (<100 FC/100 ml in 80% of samples for unrestricted irrigation) for wastewater reuse (WHO, 1973). Standards were added in the 1989 in WHO Health Guidelines for the Use of Wastewater in Agriculture and Aquaculture for wastewater irrigation of vegetables eaten raw (<1000 faecal coliforms (FC) /100 mL and <1 helminth egg /L of effluent) (WHO, 1989), based on reviews requested in 1982 by the World Bank and the World Health Organization on new epidemiological and technological evidence regarding health risks associated with wastewater irrigation (Shuval et al. 1986, Feachem et al, 1983; Strauss and Blumenthal, 1989). The new guidelines have become widely accepted by international agencies including the FAO, UNDP, UNEP and the World Bank, and have been adopted by several countries like Texas State in 1990 (75 FC/100 mL) (Papaiakovou et al., 2011), France (Ministère de la Santé et des Sports, 2010; CSHPF, 1991) and Spain ((Royal Decree 1620/2007, 2007) Iglesias et al., 2010; Iglesias Esteban and Ortega de Miguel, 2008) and number of developing, as well as developed countries. **In 2006, the World Health Organization has experienced a major turning point in its proposals with the use of quantitative microbial risk assessment** as basis for decision (WHO, 2006a,b), partly in response to criticism of too liberal previous guidelines in 1989 (WHO, 1989) compared to those of the U.S. Environmental Protection Agency (USEPA/USAID 1992). The group involved in the preparation of this third edition of guidelines has concluded that these new risk assessment studies validated the WHO (1989) of 1000 E. Coli/100ml for unrestricted irrigation of most vegetable and salad crops eaten uncooked. And Shuval et al. (2008) estimated that treating wastewater to meet the U.S.-EPA/U.S.-AID guidelines (1992) would result in an additional cost, of some $ 500,000 to $ 1,000,000 per case of disease prevented. Some countries have adopted the quantitative microbial risk assessment as a tool in their federal guidelines like Australia after 2006 (Power, 2010; EPHC/NRMMC/AHMC, 2006) and Canada (Huot, 2008). Unfortunately, the QMRA attractive approach still suffers today from lack of data and knowledge (see above). Thus, **regulations or guidelines and standards** (selected indicators associated thresholds) **greatly differ between countries**, and these differences are not easily understood without taking into account the acceptability of practices and the level of development of countries (e.g. Ongerth and Ongerth, 1982; Ongerth and Jopling 1977).

The targets of regulations (or guidelines) and standards are first to protect public health, i.e. secure wastewater reuse for agricultural product consumers as well as farmers, neighbouring inhabitants and walkers (Paranychianakis et al., 2011). They should also protect the environment – especially soils and water bodies – and agricultural productions (in quality and quantity), while environmental risks are either ignored or underrepresented (Maimon et al., 2010) and most of the standards focus on microbial health parameters, those dealing with environmental and crops being commonly not included in standards (e.g. nutrients for plants, salts, toxic organics, trace elements) (Paranychianakis et al., 2011). **Guidelines, regulations and relevant standards may also encourage wastewater reuse.** In 1970, the California State Water Code stated that *"it is the intention of the Legislature that the State undertake all possible steps to encourage development of water reclamation facilities so that reclaimed water may be available to help meet the growing water requirements of the State"* (California Water Code, section 512) and the California Water Code (section 13551) states that no one shall use water from any source of quality suitable for potable domestic use for non-potable uses, if suitable recycled water is available. Similarly, the European Communities Commission Directive declared that *"treated wastewater shall be reused whenever appropriate. Disposal routes shall* *minimize the adverse effects on the environment"* (CEC, 1991). During the last quarter of the 20th century, the benefits of promoting water reuse as a means of supplementing water resources have been recognized by most state legislatures in the USA as well as by the European Union (Asano, 2006). A first way is prohibit wastewater discharge in surface water bodies or to impose more stringent standards (Paranychianakis et al., 2011); another way is to limit the amount of conventional water use or to link permits to the progressive use of alternative water sources (Van der Bruggen, 2010). In addition, non-regulatory measures like taxes on conventional water and/or subsidies on treated wastewater and for additional infrastructures may also favour wastewater reuse. However, **inadequate regulations may prevent initiatives in wastewater reuse in irrigation**, as it has been clearly observed in Italy until 2003. In France, a similar analysis has been proposed before and after the new regulation (Ministère de la Santé et des Sports, 2010) by Lazarova and Brissaud (2007), Blin et al. (2008) and Molle et al. (2012); and a modification of this regulation being studied since about for 12 months.

Regulations or guidelines and standards on wastewater reuse in irrigation **may address wastewater treatment, the quality of treated wastewater** (chemical and biological properties) at the outlet of the wastewater treatment plant or at the point of use**, the modalities of irrigation** (crop, irrigation method, delay between the last irrigation and harvest, setback distances and/or edge required …), the traceability, the controls, the alert procedures in case of malfunction, and the public (inhabitants, education …) information. Currently, treatments and quality requirements generally depend on the crop (non-food or food crop, food crop eaten raw or cooked, edible parts in direct contact or not with irrigation water, delay between the last irrigation and harvest). They may also depend on the soil and the proximity of peoples. In only a few regulations like the French 2010 regulation (Ministère de la Santé et des Sports, 2010), the soil content in trace metal elements affect the possibility to use wastewater. Different qualities of wastewaters are generally defined. As the appropriateness of current microbial indicators are often criticised (see before), additional treatments is sometimes seen as a valuable alternative to too much controls that may be regarded as unsustainable with regard to cost.

The establishment of regulations and standards at the European scale is questionable although it is supported by several authors (Gatel, 2012; Angelakis, 2012; Angelakis 2011; Angelakis et al., 2003). Such regulations or guidelines and standards could contribute to improve the management of water resources and increase the protection of public health and environment, to avoid useless restrictions and disadvantages of national regulations (Angelakis, 2012), (iii) encourage the use of alternative water sources, and finally (iv) enhance the agricultural productivity in Southern European countries. However, regulations have to take into account actual health and environmental hazards that vary with regions and countries. Hazards caused by wastewater irrigation have to be compared to other sources of hazards, and to the cost for reducing these risks – e.g. product handling in markets (Ensink and van der Hoek, 2007) –.

The need of new matrix of multi-criteria requirements for water reuse at regional scales

Wastewater reuse practice has to insure sanitary safety, as well as economic and environmental sustainabilities **whereas scientific knowledge is still often insufficient**. Currently, there are inconsistencies in the rationale of current standards to control hazards in wastewater reuse (Salgot et al., 2006): namely the adequacy of control parameters (retained microbial (and sometimes chemical) indicators), a lack of definition of the appropriate sampling points, the number and periodicity of samples and analysis, and the cost of the analytical work (Salgot et al., 2006). Risks have to be quantitatively assessed, since a 'zero risk' policy may lead to excessive costs and a serious misallocation of expenditures (Shuval, 2008).

We suggest retaining water categories adapted by Salgot et al. (2006) with regard to special final uses (**Table 3**), as they distninguish 4 cateogries with greatly differing risks microbial contaminations. Such subdivisions could be refined according to other criteria, e.g. the time elapsed between the last irrigation and harvest (WHO, 2006), post-harvest treatments (WHO, 2006), and distances criteria (MSS, 2010).

Table 3: Microbial and chemical water quality categories for different types of irrigation of wastewater reused (from CEDEX (1997) adapted by Salgot et al. (2006))

Microbial category	Chemical category	Specific final use
I	1	• Private garden irrigation.
II	1	• Irrigation of open access landscape areas (parks, golf courses, sport fields, etc.). • Irrigation of greenhouse crops. • Irrigation of raw consumed food crops. Fruit trees sprinkler irrigated. • Unrestricted irrigation.
IV	1	• Irrigation of pasture. • Irrigation of indusrial crops for canning industry and crops not raw-consummed. • Irrigation of fruit-trees except by sprinkling. • Irrigation of industrial crops, nurseries, fodder, cereals and oleaginous seeds.
V	1	• Irrigation of forested areas, landscape areas and restricted access areas. Forestry.

Dealing with microbial hazards for humans, the detection of human pathogens may be costly, the maximum tolerable concentrations may be very low with regard to detection thresholds for the most sensitive methods (especially for viruses (WHO, 2006)), and the time needed to produce results may be long. One has therefore to propose solutions that combine (i) preventive risk management concepts (especially Hazard Analysis Critical Control Point (HACCP)) and good reuse practices to reduce the number of controls (Salgot et al., 2006), (ii) cheap and fast measurements of parameters easily monitored and partly depending on the organic load and the treatment train of wastewater (e.g. turbidity, total suspended solids, BOD, COD), (iii) fast measurements of traditional faecal indicators (*Escherichia coli* and bioindicators more resistant to treatments: e.g. *Enterococcus faecalis*, spores of *Clostridium perfringens*, viruses (bacteriophage), and (iv) maybe during high risk periods the enumeration of some actual human pathogens in raw wastewaters, especially 1 or 2 viruses (*Noroviruses*, *Hepatitis A viruses*, *Enteroviruses*, *Adenoviruses* and/or *Rotaviruses*) and protozoa (*Giardia Lamblia* cysts and *Cryptosporidium parvum* oocysts), although it would be better to measure water-based pathogens at the point of use (*Legionella*, Naegleria fowleri). Epidemiological methods seem not to be good tools for assessing health risks, since they are not sensitive enough to "tease out" cases that might be associated with recycled water. Threshold values in new standards should be based on quantitative microbial risk assessments and cost-benefit analysis insuring the economic sustainability of reuse and the best use of money. Thresholds should not to be the same between regions and countries; moreover, standards should differ between products for the local market and for export.

Dealing with soil and crop protection, some parameters may be monitored easily before irrigation in lagoons or storage reservoirs (pH, electrical conductivity), and other ones may be measured sometimes (sodium absorption ratio (SAR)). Mineral compounds that may be toxic for plants (chloride, sodium, boron) should be measured rarely as their concentration in wastewater is probably nearly constant. As long as wastewaters are of domestic origin, we suggest to check (maybe once a year or every 2 years) for the accumulation of heavy metals in soils rather than measuring their concentrations in treated wastewater, since most of them are adsorbed on microbial sludge.

By contrast, difficulties inherent in emerging organic substances result partly to their high number and diversity. Two complementary approaches may be proposed. The first is to monitor concentrations of some compounds in treated effluents; they have to be minimized with respect to the origin of the sewage (domestic, urban and/or industrial) and cover a broad spectrum of chemicals. The second is the use of bioassays on the oestrogenic (and other) activities of all compounds in treated effluents (Richard et al., 2014; Körner et al., 2001).

If microbial and chemical standards should not be the same everywhere, methods to define them could be shared. They may be based on the use of a decision support system combining a model describing the environmental fate of contaminants, a quantitative risk assessment and cost-benefit approach, all interacting with decision support systems for the optimization treatment trains in wastewater treatment plants.

Conclusions

Wastewater reuse for crop irrigation may simultaneously address water quantity and quality problems; it is implemented in regions where conventional water resources are too limiting and/or the discharge of (treated) wastewater has too much impact on the environment, especially in coastal areas and Islands where tourism is of first concern (Fazio et al., 2013). While wastewater reuse is already important in some countries/states including California (Van der Bruggen, 2010), Israel (Tal et al., 2006), and Cyprus (Papaiacovou et al., 2013), **it remains generally low**. In Europe, wastewater is preferentially reused for crop irrigation in South European countries having a high Water Stress Index, high water needs for crops and large volumes of wastewater produced (Cyprus, Malta, Spain and Italy), and reuse will increase further in these countries, even in Cyprus where it is currently limited by the collect and treatment of wastewaters (Papaiacovou et al., 2013). Wastewater reuse remains low or negligible in South European countries having a lower water stress index (Greece, France and Portugal), but it should increase because of global warming and the increase in frequency of extreme droughts. In more Northern European countries where water deficit for crops is lower or non-existent, wastewater may be reused locally for irrigation (e.g. in Germany) and/or in other sectors such as urban and industry sectors (e.g. in Belgium (Van der Bruggen, 2010)); and several large cities and conurbations depend on recharging surface water and groundwater bodies by treated wastewater, leading *de facto* to indirect potable reuse, although it is usually not acknowledged. We have obtained nearly no information on wastewater reuse in Bulgaria that has one of the highest water stress index of European countries.

Wastewater may be reused for various activities: agricultural or landscape irrigation, industrial uses, urban reuse, recreational uses, aquifer recharge, and indirect or direct potable reuse. **The type of use has to be discussed** with regard to local water needs, cost-benefit considerations and possible conflicts. Conflicts may result from competing uses in countries with a high water demand – e.g. between irrigation and industrial uses in Japan (Van der Bruggen, 2010) –, and from greatly differing quality needed for several simultaneous uses and the allocation of treatment costs – e.g. in the South of Valencia, Spain, where water from El Pindo wastewater treatment plant simultaneously irrigates submerged rice fields and supplies Albefura lake (Renault et al., 2013) –. **Public acceptance is generally good for reuse in irrigation** and for other uses, with the exception of direct potable reuse (Ormerod and Scott, 2012; Cain, 2011; Leverenz et al., 2011; Sofres, 2006; Ongerth and Ongerth, 1982; Ongerth and Jopling, 1977) although some 'success stories' exist in direct potable reuse as in Windhoek (Namibia) (du Pisani, 2006). However, **some opposition exists**. They are often explained by the 'yuck factor' (Macpherson, 2013; Leong, 2010), corresponding to a psychological aversion with the following expressions retained in surveys: "psychologically repugnant", "lack of purity", "can cause disease" (Ongerth and Jopling, 1977). Another explaination is that public opposition would result from social and cultural perceptions of risk (Ormerod and Scott, 2012). The 2 preceding explanations **justify more information** dealing with water cycle, water treatments and the actual risks **in order to prevent project failure**.

Actual risks include sanitary, environmental and agricultural hazards that result from the presence in raw sewage of human pathogens and various inorganic and organic compounds. Although it is possible to produce water of almost any quality desired from wastewater (Norton-Brandão et al., 2013), cost-effectiveness of treatments have to be ensured. The management of conventional and alternative water resources requires appropriate regulations and standards, as well as economic policy. While the diversity of current rules between European countries seems not scientifically justified and leads to inequalities, differences between regions in water requirements, raw wastewater properties and human resistance to pathogens justify having regulations adapted to regional contexts, and Ashbolt et al. (2001) to tailor indicator choice to local circumstances when translating international guidelines into local standards. **The European Union could propose guidelines with maximum tolerated risks and a methodological framework to elaborate regional or national regulations and standards** that account for local specificities, in the same spirit that the World Health Organization encourages national governments to adapt their guidelines to their own socioeconomic and environmental realities (Ensink and van der Hoek, 2007). A distinction should then be performed between crops for local markets or for export.

Some separate tools exist to optimise wastewater treatment, assess quantitatively microbial risks or estimate the balance between benefits and costs, including a monetary valuation of environmental changes, but **new tools combining risk assessment, treatment optimisation and cost-benefit considerations are required to support decisions** dealing with the definition of new regulations and standards as well as economic policy (tariffs, taxes, subsidies …). In the current state of knowledge, **a first generation of combined tools can be proposed, but they would have to evolve** in order to (i) incorporate new data, processes and pathways of contamination, (ii) add emerging microbial or chemical contaminants, and (iii) adapt to possible changes in standards (indicators themselves, their maximum tolerable quantities and/or their minimum removal rate).

The relevance of microbial indicators has to be discussed. Their use to characterize the efficiency of treatments is probably relevant as long as several bioindicators having different sensitivities to different treatments are used simultaneously. **The major limitations of microbial indicators** generally used **are first weak correlations with the level of pathogens** in wastewater **and second their fate in the environment that may differ from the fate of pathogens**. In particular, they are generally not correlated with the levels of enteric viruses that are increasingly considered as the etiologic agents of human enteric infections. **An alternative could be to detect directly some pathogens** (Bosch et al., 2008) from a list established using a methodology as that of the U.S. Environmental Protection Agency and the American Water Works Association. This choice is currently hampered by the low levels of enteric pathogens and the very low thresholds of tolerable concentrations; they may require the concentration of water samples prior to pathogen detection and the use of molecular methods. But great technological progresses have been performed, including those using DNA microarrays on silicone nanostructures (e.g. Oillic et al., 2007), and molecular methods would allow to obtain results quickly. **Another alternative would be to partly replace microbial controls with more**

advanced treatments and/or more controls on some treatments (e.g. by measuring the residual content in disinfectant), all the more that treatments will probably have to become more stringent, partly to avoid environmental discharge of water too much contaminated by chemicals. In the case of separate sewage networks for domestic wastewaters and rain runoff without industrial wastewaters, **indicators of emerging organic pollutants have probably to be chosen** among chemicals from personal care products, pharmaceutical products, bisphenol A and phthalates; otherwise they must cover a broad spectrum of toxicological and ecological risks as well as possible technical disorders (Salgot et al., 2006).

Additional knowledge is needed to optimize wastewater reuse. **First, it is important to better understand and describe the processes implied in microbial contaminations** in order to improve quantitative microbial risk assessment: ***via* air** (aerosolization, atmospheric speciation of pathogens, inactivation/mortality and transport in the atmosphere, redeposition, transfer from human respiratory tract to gastro-intestinal track), ***via* food** (internalisation through the roots, the leaf stomata and/or injuries in the aerial parts of plants), **and *via* water to some extent** to better assess the reactive transport of pathogens from soil surface to underlying aquifer. **It is also important to assess the combined effects of organic pollutants on human health and environmental biodiversity, as well as their cumulative consequences over long-term periods**, as concentrations of most of them in urban wastewaters are generally below the toxic levels for humans. Quantitative microbial risk assessment and quantitative chemical risk assessment need also informations on population habits (food, displacements, protection of farmers during field labour …) that are probably not available in each European country. **Second, new probe methodologies are required** to monitor *in situ* microbial and chemical contaminations in real time and at low costs, whereas current cost greatly vary between microorganisms, as well as between organic micro-pollutants (Salgot et al., 2006). **Third, the monetary valuation of the changes resulting from wastewater reuse** (human health, environment, recreational activities, industry, jobs …) must be refined, especially the monetary valuation of environmental changes, which is recent and may result from different calculation procedures.

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Appendix 1: Virus fate in the environment

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Viruses are obligate, intracellular pathogens (Fong and Lipp, 2005; Sobsey and Meschke, 2003). Outside of the cells they may infect, virus particles (virions) consist of the genetic material (viral RNA or DNA) wrapped by a protein coat (viral capside) and, in some cases, an external envelope of lipids that surrounds the capside. **Most of human enteric viruses are naked viruses**, i.e. without lipidic coating (Fong and Lipp, 2005); they are then very resistant in the environment and to certain water treatments (Carducci et al., 2009), and **they may survive a long time in water environments** (weeks to months) (La Rosa et al., 2012). An exception is the well-known severe acute respiratory syndrome (SARS) virus, which is an enveloped virus, i.e. with a lipidic coating, that may be considered as enteric and that is rapidly inactivated in water and wastewater at ambient temperatures (Gundy et al., 2009; Bennett, 2006). **Most of human enteric viruses are human specific**, except hepatitis E virus with infections from pigs suggested from almost identical viruses (Koopmans and Duizer, 2004) and infection proven from deer (Tei et al., 2003), and the severe acute respiratory syndrome virus which moves from the bat population to other animals and humans (Gundy et al., 2009; Bennett, 2006). **The most notable viruses are hepatitis A virus** (HAV) (hepatitis, liver damage and jaundice), and the *Calicivirus*, especially the ***Norovirus*** genus (diarrhoea), and in a lesser extent the ***Rotavirus*** (diarrhoea) (Rodríguez-Lázaro et al., 2012; Carducci et al., 2009; Koopmans and Duizer, 2004). Other viruses of greatest concern in water (and their associative illnesses) include the group of ***Enteroviruses*** with *Poliovirus*, *Coxsackievirus* and *Echovirus* (diarrhoea, meningitis, myocarditis, fever, respiratory disease, nervous system disorders, birth defects), ***Astrovirus*** (diarrhoea) and ***Adenovirus*** (diarrhoea, respiratory disease, eye infections. heart disease) (Rodríguez-Lázaro et al., 2012). They have been involved in waterborne outbreaks *via* drinking water (Hoffman et al., 2009; Zhuang and Jin, 2008; Craun et al., 2006; Gerba, 1999) and recreational waters (swimming pools, lakes, lagoons, rivers and thermal station) (Sinclair et al., 2009; Calderon et al., 2005; Craun et al., 2005). The most commonly reported foodborne viral infections lead to gastroenteritis and less frequently hepatitis A (Seymour and Appleton, 2001). *Norovirus, Enteroviruses* (*Coxsackievirus* and *Echovirus*) and hepatitis A virus are among the 12 human pathogens listed in the 'Contaminant Candidate List' established by the U.S. Environmental Protection Agency in 2008 (US-EPA, 2009), and among the 11 human pathogens retained nearly at the same time by the American Water Works Association (AWWA) (Hoffman et al., 2009); In addition, *Adenovirus* and *Poliovirus* are in the first list, whereas *Rotavirus* is in the second one. The hepatitis A virus and the *Norovirus* have been detected in wastewaters at the entrance and the exit of wastewater treatment plants (Da Silva et al., 2011; La Rosa et al., 2010; Petrinca et al., 2009; Van den Berg et al., 2005; Pusch et al., 2005). They have been also detected sometimes in soils and underground waters (Parashar et al., 2011; Reynolds et al., 2008; Borchardt et al., 2003).

The epidemiology of the Hepatitis A virus varies with the public hygiene and the water treatment. There are endemic zones in Africa (Kamal et al., 2010; Gharbi-Khelifi et al., 2006) and in some European countries like Albania (Divizia et al., 2005), where human populations are exposed to this virus from childhood; viral infections are then generally asymptomatic or lead to not acute disease (Yong and Son, 2009; Pinto et al., 2007). In regions like Tunisia, hepatitis A virus infections seem to be cyclic with maximum in winters that may result from heavy winter rainfalls, lower temperature than in summer, the use of unsanitised sludge as crop fertilizer, and higher (infected) shellfish consumption (Gharbi-Khelifi et al., 2006). With the development of this country, asymptomatic infections in young children have tended to decrease, and more severe disease with even deadly forms of infection have tended to appear among adults, clinical manifestations seeming to increase with age (Gharbi-Khelifi et al., 2006). In regions with higher public health, viral infections result first from the Hepatitis A virus importation from endemic regions *via* travellers or foods that are eaten crude or only slightly cooked (seashells, onions, lettuce, spinach, tomatoes, raspberries …) (Hollinger and Emerson, 2007); human adults then develop acute forms of the disease, see rarely fulminant forms (Yong and Son, 2009; Pinto et al., 2007). Two hepatitis A outbreaks affecting more than 800 individuals were reported in western France (between December 1992/March 1993 (Nuiaouet et al., 1993) and in the Italian lake district between November 2002 and February 2003 (Divizia et al., 2005). The great genetic and antigenic variabilities of the *Norovirus*, and their dynamic behaviour render difficult their detection and characterization (Green, 2007) and don't enable humans to immunize themselves (Atmar, 2010; Green, 2007). Gastroenteritis resulting from *Norovirus* infections (also called ‘winter vomiting disease’) are generally not severe. They often combine diarrhea and vomiting, and affect all the age groups (Green, 2007). They explain in several developed countries more than 80% of gastroenteritis outbreaks for adults (Koopmans et al., 2000) and more than 90% of nonbacterial outbreaks (Green, 2007). Several contaminated foods are incriminated (Green, 2007; Koopmans and Duizer, 2004). Infections vary with seasons (Yen et al., 2011; Chikhi-Brachet et al., 2002) although this seasonality is not necessarily found for virus in raw and treated wastewaters (Myrmel et al., 2006; Van den Berg et al., 2005).

Viruses have the greatest infectivity of all waterborne microorganisms: a few number of them (about 10^1-10^2 pfu) being sufficient to cause infection (Morin and Picoche, 2008; Hollinger and Emerson, 2007; Green, 2007), **while they are excreted in the faeces in the largest numbers**, i.e. up to 10^{11}-$10^{12}.g^{-1}$ faeces for sick and healthy carriers (Maunula et al., 2013; La Rosa et al., 2012; Da Silva et al., 2011; Reynolds et al., 2008; Fong and Lipp, 2005; Koopmans and Duizer, 2004) and several weeks after the illness period (Maunula et al., 2013; Da Silva et al., 2007). In the USA, 8.2% of drinking waterborne outbreaks recorded between 1911 and 2006 had a well identified viral origin and other 44.6% corresponded to acute gastroenteritis with an undetermined etiology (Craun et al., 2010) that were compatible with viral infections (Reynolds et al., 2008). In France, some foodborne outbreaks resulting from water contamination by viruses were noted in the city of Gourdon (Lot) in August 2000 (INVS, 2001) and in Isère department in November 2002 (INVS, 2004).

Viruses may be transmitted by direct contamination (faecal-oral) **or indirect contamination** *via* contaminated environmental surfaces (Vasickova et al., 2010; Barker et al., 2004) or more generally *via* contaminated food or drinkable water (Vega et al., 2005; Koopmans and Duizer, 2004; Pallansch, 2001) after transport in the environment (surface, waters, soils). Transmission *via* air has also been noted (Ziros et al., 2011; Marks et al., 2000; Caul, 1994). The transmission of enteric viruses present in wastewaters *via* the environment has been neglected for a long time with regard to their direct transmission (faecal-oral), and with regard to other pathogen transmission (bacteria and parasites like protozoa and helminths) for which detection/enumeration methods exist since more time (Koopmans and Duizer, 2004). With the recent development of detection methods based on molecular biology that are very sensitive and that may be applied on environmental samples, the environmenntal transmission of human viruses has become an emerging problematic (Rodríguez-Lázaro et al., 2012; Cliver, 2010; Carducci et al., 2009; Costafreda et al., 2006; Widdowson et al., 2005; Brooks et al., 2005; Fong and Lipp, 2005; Bhattacharya et al., 2004; Koopmans and Duizer, 2004; Metcalf et al., 1995) and human viruses are more and more often incriminated in human outbreaks (Koopmans and Duizer, 2004; Gerba, 1999).

Human enteric viruses are not efficiently removed by conventional filtration and they are more resistant to disinfectants than bacteria (CFPTEP, 2010a; CFPTEP, 2010b; Aronino et al., 2009; Petrinca et al., 2009; Da Silva et al., 2008; Koopmans and Duizer, 2004), although some studies have shown the high efficiency of some treatments – e.g. the removal of *Norovirus* with differences in the decrease of GI and GII *Norovirus* groups (Da Silva et al., 2007), and the nearly complete removal of viruses by the treatment train in the Upper Occoquan Sewage Authority Water Reclamation Plant that protect the Occoquan Reservoir (Rose et al., 2001) –. Viruses are detected in raw wastewaters as well as in treated wastewaters at the exit of wastewater treatment plants – e.g. for 5 wastewater treatment plants around Rome (La Rosa et al., 2010), for 2 wastewater treatment plants in the southwest of the Netherlands (Van den Berg et al., 2005), for the wastewater treatment plant of Leipzig (Pusch et al., 2005), for Rio de Janeiro in Brazil (Villar et al., 2007), and for Oslo area and elsewhere in Norway (Myrmel et al., 2006) –. This may partly explain why they are also found in conventional surface waters and groundwaters. Because of their small size (0.02-0.1 µm) and ease of transport in the subsurface, viruses are of primary concern in groundwaters (Reynolds et al., 2008).

Supplied during irrigation by wastewaters, human enteric viruses may be dispersed in the atmosphere as bioaerosols, **or transported within the soil towards the aquifers** (Zhuang and Jin, 2008; Reynolds et al., 2008; Gerba, 1999) **and other environmental reservoirs** (surface water bodies, roots of plants …). The hepatitis A virus and the *Norovirus* may be absorbed and internalised in the roots of onions and lettuce and are thereafter recovered in their bulb and leaves, respectively (Wei et al., 2011; Urbanucci et al., 2009; Wei et al., 2009; Chancellor et al., 2006). **In the soil, virus fate depends on their transport, their adsorption on solids** (Da Silva et al., 2011; Syngouna and Chrysikopoulos, 2010; Zhuang and

Jin, 2008; Guan et al., 2003; Dowd et al., 1998; Vilker et al., 1983; Moore et al., 1981; Taylor et al., 1981)**, and their inactivation** (i.e. either their disappearance or the loss of their infection potential) (Murray and Laband, 1979). Transport, immobilisation/mobilisation and inactivation depend on each other: virus may be transferred either free or adsorbed on a colloid (Syngouna and Chrysikopoulos, 2010; Zhuang and Jin, 2008), and virus inactivation may be slowed (Schaub and Sagik, 1975; Gerba, 1999) or accelerated by their adsorption on some minerals, especially on metal oxides (Zhuang and Jin, 2008; Murray and Laband, 1979).

We group here the **too rare works** that **have dealt with the fate of human enteric viruses in the Environment** with those that used bacteriophages as models, although there are differences between the behaviours of bacteriophages and human enteric viruses of the *Enterovirus* genus, as well as between viruses within the *Enterovirus* genus (*Coxsachievirus*, *Enterovirus*, *Poliovirus* and *Echovirus*) (Goyal and Gerba, 1979; Gerba et al., 1981).
Several studies have dealt with the adsorption of viruses on solids. **Although a few ones have focused on real soils** (Bradley et al., 2011; Taylor et al., 1981; Moore et al., 1981)**, the majority have considered pure materials** as models of solid behaviour: **metal oxides** (Zhuang and Jin, 2008; Buining et al., 1994; Murray and Laband, 1979), **clays** such as kaolinite and montmorillonite (Syngouna and Chrysikopoulos, 2010; Lipson and Stotzky, 1983; Vilker et al., 1983; Taylor et al., 1981), **silica** (Taylor et al., 1981; Murray and Laband, 1979) **and organic compounds** (Zhuang and Jin, 2003). To our knowledge, the effect of the soil structure has not been studied. Viruses that were used were bacteriophages (MS2, φX174, PRD1 …) differing from each other by their dimension (Dowd et al., 1998), their isoelectric point (Michen and Graule, 2010), their wettability (Dowd et al., 1998) and residues of amino-acids on the surface of the capsid (Syngouna and Chrysikopoulos, 2010). A few actual human pathogens have been used: the *Poliovirus* (Taylor et al., 1981; Moore et al., 1981; Murray and Laband, 1979) and other viruses also belonging to the *Enterovirus* genus (*Enterovirus*, *Coxsachievirus*, *Echovirus*) (Gerba, 1999), the possibility to cultivate them *in vitro* permitting to assess their infectious behaviour. Some alternatives to the use of human pathogen viruses have emerged. On the one hand, animal viruses have been used as surrogates of human viruses: the murine *Mengovirus* as a surrogate of the hepatitis A virus (Costafreda et al., 2006), the feline *Calicivirus* as a surrogate of the human *Norovirus* (Park et al., 2011; Bae and Schwab, 2008), and more recently, the murine *Norovirus* as another surrogate of the human *Norovirus* (Park et al., 2011; Hewitt et al., 2009; Bae and Schwab, 2008). On the other hand, virus-like particles (VLP) quite similar to actual viruses may be constructed for some viruses by synthetizing *in vitro* the capsidal recombinant proteins issued from the same virus that self-assemble: this is the case for the *Norovirus* (Da Silva et al., 2011; Goodridge et al., 2004) and the *Rotavirus*, although the actual structure of *Rotavirus* capside is more complex and is not perfectly reproduced by virus-like particles (Charpilienne et al., 2001). Temperature effects have been studied only a little, whereas these effects indirectly inform on the hydrophilic/hydrophobic forces involved in adsorption (Syngouna and Chrysikopoulos, 2010) and climate affects *Norovirus* epidemiology (Lopman et al., 2009). Studies on virus immobilisation have often focused on equilibrium reached after periods of about 1 hour (Moore et al., 1981) or some days (Syngouna and Chrysikopoulos, 2010), but recent works define the attachment efficiency as the actual to maximal absorption (in non-limiting conditions) rate ratio (Da Silva et al., 2011). Only a few

studies have dealt with the reversibility of the immobilisation (Zhuang and Jin, 2008; Loveland and al., 1996; Murray and Laband, 1979). Murray and Laband (1979) followed the remobilisation of intact (infectious) *Poliovirus*, the proteins that are constitutive of their capsid being labelled with ^{14}C and their RNA being labelled with ^{3}H; they assessed the implication of the physicochemical denaturation on virus inactivation. Some mathematical models have been proposed, including models that account for spatial heterogeneities in advection-convection, geochemistry and inactivation in solution and at solid surface (Tufenkji, 2007; Bhattacharjee et al., 2002; Schijven and Hassanizadeh, 2000). And some studies compared experimental data and model simulations, but model hypotheses could be criticised (Zhuang and Jin, 2008; Guan et al., 2003; Chu et al., 2001). Empirical models also exist for virus inactivation in other matrix and extreme conditions with regard to soil classical ones, like hepatitis A virus in acidified berries (Deboosere et al., 2010) and hepatitis A virus and *Norovirus* on inert solid surfaces (Kim et al., 2012).

Virus immobilisation depends (i) on the virus itself (Syngouna and Chrysikopoulos, 2010; Michen and Graule, 2010; Zhuang and Jin, 2008; Goodridge et al., 2004; Guan et al., 2003; Dowd et al., 1998; Gerba et al., 1981; Goyal and Gerba, 1979; Chlumecka et al., 1977), the viral strain for the *Norovirus* (Da Silva et al., 2011) or a change in the virus conformation under certain circumstances for the *Poliovirus* (Taylor et al., 1981), (ii) on soil solid nature (Bradley et al., 2011; Zhuang and Jin, 2008; Zhuang and Jin, 2003; Moore et al., 1981; Murray and Laband, 1979), and (iii) on the soil solution properties (pH, ionic strength, mineral ions, organic compounds) (Da Silva et al., 2011; Cao et al., 2010; Zhuang and Jin, 2003; Dowd et al., 1998; Taylor et al., 1981). Forces involved in virus adsorption are of various natures. They include electrical forces (Schaldach et al., 2006), the virus and soil inert particles charges varying with the pH and the ionic composition of the solution (Cao et al., 2010; Van Voorthuizen et al., 2001; Sposito, 1998; Chorover and Sposito, 1995; Gerba et al., 1981); they are attractive when the pH is between the isoelectric point of the virus and the isoelectric point of the soil particle (Goyal, 1979), the retention intensity being then often inversely proportional to the pH (Syngouna and Chrysikopoulos, 2010). The salinity may affect (Cao et al., 2010), and organic compounds may counter act the effect of an increase of salinity (Cao et al., 2010). A few solids, like Fe oxides, have high isoelectric points that favour virus adsorption over a larger range of pH (Syngouna and Chrysikopoulos, 2010). However, adsorption may also exist if electric repulsion is reduced by the presence of divalent cations (Ca^{2+}, Mg^{2+}) (Da Silva et al., 2011) or an increase in the ionic strength of the solution (Sposito, 1998), and/or when other forces counterbalance this repulsion: van der Waals forces (Chattopadhyay and Puls, 2000), hydrophilic/hydrophobic attractions (Van Voorthuizen et al., 2001) …. Some anions also affect virus immobilisation: Cl^- seems to be the most favourable to virus immobilisation, HPO_4^{2-} and HCO_3^- may favour or disfavour virus immobilization depending on the virus itself and the physicochemical conditions (Da Silva et al., 2011). Forces involved in colloid adsorption may also include hydrophobic/hydrophilic forces (Crist et al., 2005). DLVO theory (Derjaguin, Landau, Verwey et Overbeek) describe some of these forces but ignore other forces that may be sometimes more important (Attinti et al., 2010; Syngouna and Chrysikopoulos, 2010) and ignore the physical retention of the larger viruses (Dowd et al., 1998) and of viral aggregates in conditions favouring virus aggregation (Da Silva et al., 2011). The inability of the DLVO theory (Hermansson, 1999) to model the retention of some bacteria suggests that processes are ignored, including clogging of the pores and retention by particle roughness (Jacobs et al., 2007). Virus adsorption at the air-water interface may be taken into account in unsaturated systems (Chu et al., 2001), as also noted for bacteria (Schäfer et al., 1998).

Only a few works have dealt with virus inactivation in soils (Zhao et al., 2008) and few is known on their survival and inactivation (Rzeżutka and Cook, 2004; Gerba, 1999). **Virus adsorption on solids can slow** (Schaub and Sagik, 1975; Gerba, 1999)**, or accelerate virus inactivatio**n. **Virus inactivation at the surface of metal oxides can be fast** (Zhuang and Jin, 2008; Murray and Laband, 1979); it may be accelerated by factors favouring indirectly virus adsorption, including a decrease of soil moisture and soil sterilization (Zhao et al., 2008), a decrease in soil solution pH (Zhuang and Jin, 2008) and a decrease in other compounds that may be adsorbed at the surface of oxides (e.g. phosphates (Zhuang and Jin, 2008)); by contrasts, compounds may may favor specifc attachments and inhance virus inactivation (e.g. carbonate (Zhuang and Jin, 2008)). Virus inactivation has been studied in other matrix and in extreme conditions with regard to those prevailing in soils, including the inactivation of hepatitis A virus in acidified berries at high temperatures (Deboosere et al. 2010), and more generally virus inactivation in foods by traditional and novel technologies (Hirneisen et al., 2010). Various processes may explain viral inactivation: viral RNA or DNA damage (with or without preliminary release of viral RNA or DNA from the capside; virus antigen damage, separation between viral DNA or RNA and the capsid; in special laboratory contexts Poliovirus RNA separated from the capsid may still infect host cells (Nuanualsuwan and Cliver, 2003).

By contrast, **nearly nothing is known on virus fate in the atmosphere**. Studies on atmospheric fluxes of biocolloids have begun only recently and enumerating the specific content in bacteria and viruses remain difficult (Georgakopoulos et al., 2009; Georgakopoulos et al., 2008; Verreault et al., 2008). Some studies deal with airborne transmission of viruses in hospital (Caul, 1994), and in the vicinity of wastewater treatment plants (Ziros et al., 2011). Several factors affecting survival and mobility are known or greatly suspected: (i) the relative humidity and temperature (Kim et al., 2012; Donaldson, 1972; Akers and Hatch, 1968; De Jong and Winkler, 1968; Akers et al., 1966), (ii) sunlight or UV radiations (Park et al., 2011; Simonet and Gantzer, 2006b; Thurston-Enriquez et al., 2003; Sinton et al., 2002; Fujioka and Yoneyama, 2002), and (iii) oxidants (ozone (Tseng and Li, 2006), free radicals OH• …). Inactivation by solar UV radiation of bacteriophage and polioviruses in marine water reported by Fong and Lipp (2005), and inactivation of adenovirus, coliphage MS-2, and feline calicivirus in buffered demand-free (BDF) water and groundwater (Thurston-Enriquez et al., 2003); however, they noticed that viruses are more resilient than many other pathogens to UV radiations and that viruses with double-strand DNA or RNA are extremely stable when exposed to UV because their undamaged DNA or RNA strand may serve as a template for repair by host enzymes (Thurston-Enriquez et al., 2003; Gerba et al., 2002).

Difficulties in virus detection and enumeration result first from their low infectious doses (10^1-10^2 viruses) (Morin and Picoche, 2008; Hollinger and Emerson, 2007; Green, 2007)**, and the very low tolerable concentration threshold** – e.g. 5 10^{-3} *Rotavirus*/L to insure less than 10^{-6} Disability Adjusted Life Years (DALY) per person per year (pppy) (WHO, 2006) –. **Additional difficulties result from the extraction of viruses** from solid matrix (soil, food …) that are always partial, and **the enumeration of infectious viruses that remains impossible for viruses not yet culturable** – e.g. *Norovirus* –. The overall method to extract and detect viruses could be divided into three different steps: (1) virus elution and clarification from substrates, (2) virus concentration (Hamza et al., 2009; Croci et al., 2008; Liu et al., 2007; Villar et al., 2006; Dubois et al., 2006; Katayama et al., 2002; Jothikumar et al. 1995; Tsai et al., 1993), and (3) virus enumeration. For each of these steps various methods exist. Virus enumeration has greatly evolved during the last decades with molecular biology (PCR, RT-PCR, quantitative RT-PCR …) (Mattison and Bidawid, 2009; Villar et al., 2007; Da Silva et al., 2007; Costafreda et al. 2006; Brooks et al., 2005; Bhattacharya et al., 2004; Metcalf et al., 1995). In the environment, virus enumeration depends simultaneously on their elution and extraction from solids and their subsequent concentration which may lead to simultaneously concentrate PCR inhibitors in some methods (Hamza et al., 2009; Croci et al., 2008; Dubois et al. 2006). Cultivation methods are fastidious, and require the availability of target cells, fast enough virus replication and observable cytopathic effects. As it is difficult to estimate the proportion of viruses that remain infectious (Nuanualsuwan and Cliver, 2002), some PCR methods have been proposed to discriminate between infectious and non-infectious viruses (Bhattacharya et al., 2004; Nuanualsuwan and Cliver, 2002). However, it depend on the mode of inactivation of viruses and their effect on viral RNA or DNA (Simonet and Gantzer, 2006a). Other methods of detection and enumeration exist like immunochromatographic assays for the detection of human *Rotavirus* (Bon et al., 2006), Characterization and purification of viruses using chromatofocusing applied on bacteriophages (Brorson et al., 2008).

Appendix 2: Organic contaminants in wastewaters

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Introduction: main sources of organic contaminants in wastewaters

Organic contaminants (OC) in wastewaters come from several sources. We mainly focus on OC in domestic wastewaters, although wastewater treatment plants (WWTP) often receive a mixture of domestic and industrial waters, as well as runoff waters during rainy events. As a consequence of the diversity and complexity of OC, there is an abundant literature but only a few general papers, while others deal with case studies that are difficult to generalize since they depend on the experimental conditions and treatment trains in WWTPs.

Components of personal care products used in large quantities are widely found in raw wastewaters (bath additives, shampoos, hair tonic, skin care products, hair sprays, soaps, sun screens, perfumes, aftershaves …). Other compounds issued from personal care products and found in sewage include parabens (alkyl-*p*-hydroxybenzoates) that are among the most widely used antimicrobial preservative (for cosmetics, toiletries, pharmaceuticals and even foodstuffs), the triclosan (a chlorinated diphenyl ether) used as antiseptic agent and preservative (for toothpaste, footwear, handsoap, acne creams …), the byphenylol and the chlorophene also used as preservatives and disinfectants. Pharmaceutical products most commonly detected in the treated effluents worldwide include antibiotics, lipid regulator agents, anti-inflammatory drugs, ß-blockers, cancer therapeutics, contraceptives and other hormones (Hernando et al., 2006; Nikolaou et al., 2007). Some pharmaceuticals and personal care products are very persistent in the environment (e.g. blood lipid regulators such as clofibric acid, X-ray contrast media and musks). **Phthalates are other ubiquitous OC in the environment**. They have been used for 50 years and 3 million tons are produced per year around the world. They are present in many products, for examples in plastics (e.g. PVC) as plasticizers to make them flexible and improve both impact and cold resistances, and in cosmetics as fixative agents to increase the penetrating power of a product into the skin or to prevent cracking of nails. The most used phthalate is DEHP (di-2-ethylhexyl phthalate) especially for fragrances, food containers, blood bags, catheters or bibbers; it is also the most quantified in wastewater (Barnabé et al., 2008; Dargnat et al., 2009; Clara et al., 2010). The concentration in wastewater varied from 10 to 200 µg/L (Fromme et al., 2002; Vogelsang et al., 2006; Gasperi et al., 2008a). **Pesticides are also found in wastewaters**. Although the largest amounts are used for agriculture, they are also used in urban areas to protect infrastructures (buildings, roads, streets, railway tracks, gardens …). When used in urban areas, especially on impermeable surfaces, pesticides may move into sewers during runoff events. Blanchoud et al. (2007) revealed that 80% to 100% of the diuron applied on impervious surfaces could potentially be remobilized during a rainfall event. These authors estimated also than the urban and agriculture wastewaters contribution to the pesticide water pollution was equivalent in Marne watershed. Others biocids are widely used in urban areas for material protection. Depending on the existence of combined or separate urban sewer networks for house wastewater and rainwater, pesticide can directly contaminate or not wastewaters (Gerecke et al., 2002). Gasperi et al. (2008b) identified the origin of OC and metals into the sewerage system of Paris. Of the 66 elements investigated (based on the list established in European Decision no. 2455/2001/EC), 33 and 40 priority substances could be observed in raw sewage and wet weather effluents, respectively. Chlorobenzenes and most of the pesticides always remained below the limit of quantification, while the majority of other OC assessed were identified within the µg/L range (Gasperi et al., 2008b). Runoff via atmospheric inputs and/or surface leaching was found to induce a wider range of OC and lead to higher concentrations of certain PAHs (3–4 rings PAHs), pesticides (diuron and oxadiazon, and to a lesser extent, of diazinon, propiconazole and terbutryn) and organotin compounds (Gasperi et al., 2008b). **PAHs found in wastewater are indicators of pyrolytic inputs** (mainly coming from the use of fuels in house heating and transport). PAH concentrations reported in the literature for domestic wastewaters ranged between 0.02 to 0.89 µg/L (Palmquist and Hanaeus, 2005). Other ubiquous persistent organic pollutants are the **PCBs**; however, articles concerning their concentrations in wastewater are the rarest (Miège et al., 2009).

Glassmeyer et al. (2005) tried to regroup statistically OC depending on their occurrence and detection frequencies. **OC with similar use were frequently grouped together**. For example, the pharmaceuticals trimethoprim, sulfamethoxazole, dehydronifedipine, diphenhydramine, diltiazem, and carbamazepine were all grouped together. Other notable groupings were (i) faecal sterols, cholesterol, coprostanol, and sitosterol, (ii) caffeine and its metabolite, and (iii) the musks tonalide and galaxolide. The most unexpected result for Glassmeyer et al. (2005) was that acetaminophen appeared grouped with the two microorganisms, *Escherichia coli* and *Enterococci*, and not the other pharmaceuticals. Compounds that are typically only used by humans, such as the pharmaceuticals carbamazepine and diphenhydramine, and even caffeine, would be potential candidates as indicators of water of human origin (Chen et al., 2002; Buerge et al., 2003; Clara et al., 2004; Glassmeyer et al., 2005). It is also the case of sterols, mainly the coprostanol (Glassmeyer et al., 2005).

Focazio et al. (2008) propose to divide OC detected in wastewater into **16 groups**: **steroids, non-prescription drugs, fragrances and flavors, antibiotics, pesticides, other prescription drugs, fire retardants, plasticizers, insect repellent, detergent metabolites, disinfectant, cosmetics, polynuclear aromatic hydrocarbons (PAH), solvents, dyes/resins/fuels and antioxidants**. Each group includes various compounds that often distinguish from each other by very different chemical structures, physico-chemical properties, and degradation products issued from the human metabolism or from biological or chemical degradation into sewers or WWTP.

The academic interest in recent decades for the presence of **pharmaceutical residues** in wastewater results in a large number of publications on this subject (Miège et al., 2009). The molecules studied are the most commonly prescribed antibiotics (ciprofloxacin, doxycyclin, norfloxacin, trimethoprim and sulfamethoxazole) and analgesics and anti-inflammatory drugs (diclofenac, ibuprofen, and naproxen) (Miège et al., 2009), the number of studies decreasing for the phthalates with DEHP and BBP, and finally bisphenol A. Molecules least cited in the literature are contrast agents, ß-blockers, lipid regulators and finally diuretics (Miège et al., 2009). We have selected some results in order to point out the main figures on the occurrence and fate of OC linked to the wastewater, and to assess potential risks related to the irrigation with wastewater. C**oncentrations of most OC in urban wastewaters** (raw or treated) **are generally below the toxic level**s for humans, but potential **problems may result from the combined effects of several pollutants and/or their cumulative consequences over long-term periods**, either directly or indirectly through modifications of ecosystems (e.g. several soil microorganisms acquiring antibiotic resistance). This last point is not treated here; it would be probably more pertinent in the case of manure or animal effluent spreading.

Organic contaminants in the environment; link with wastewaters

As wastewaters explain most the environmental dispersion of pharmaceuticals, their monitoring in the environment can give interesting informations, as highlighted in the review of Monteiro and Boxall (2010), the monitoring work of Gros et al. (2010) on 73 pharmaceuticals, and a review on the presence and persistence of pharmaceuticals in the environment, with data from more than 150 references (Glassmeyer et al., 2008).

Focazio et al. (2008) analysed U.S. surface waters and groundwaters, and detected 63 OC among 100 listed in at least 1 water sample; they assumed that OC originated from wastewaters. The maximum number of compounds detected in the same site was 31 and **the median number of compounds detected per site was 4**. In surface water, the 5 most frequently detected OC were cholesterol (59% of positive samples, natural sterol), metolachlor (53%, herbicide), cotinine (51%, nicotine metabolite), β-sitosterol (37%, natural plant sterol) and 1,7-dimethylxanthine (27%, caffeine metabolite). In groundwater, the 5 most frequently detected chemicals were tetrachloroethylene (24%, solvent), carbamazepine (20%, pharmaceutical), bisphenol A (20%, plasticizer), 1,7-dimethylxanthine (16%, caffeine metabolite) and tri(2-chloroethyl)phosphate (12%, fire retardant). Non-prescription drugs (including caffeine, 1,7-dimethylxanthine, cotinine, ibuprofen and acetominophen) were detected more frequently than any other of the 15 groups in surface water; only 3 groups (biogenic steroids, detergent metabolites, and solvents) had individual chemical maximum concentrations exceeding 2 µg/L, whereas 7 groups (including antibiotics, non-prescription drugs, and other prescription drugs) had maximum concentrations lower than 0.5 µg/L.

Another interesting review by Deblonde et al. (2011) mainly focused on bisphenol A, 6 phthalates and 50 pharmaceuticals (including drugs for human health and disinfectants). The molecules studied were the most commonly prescribed antibiotics (ciprofloxacin, doxycyclin, norfloxacin, trimethoprim and sulfamethoxazole), analgesics and anti-inflammatory drugs (diclofenac, ibuprofen, and naproxen), whereas the least cited molecules are contrast agents, ß-blockers, lipid regulators and diuretics (Miège et al., 2009). In the review of Deblonde et al. (2011), the concentrations of OC in the influent of WWTPs ranged from 0.007 to 56.63 µg/L. Caffeine concentration in influent was the highest among the concentrations of molecules investigated (average of 56.63 µg/L). The concentrations of ofloxacin were the lowest, and varied between 0.007 and 2.275 µg/L in the influent treatment plant. Tetracycline, ibuprofen, contrast products, caffeine, and codeine were found in effluents of WWTPs (Deblonde et al., 2011), also phthalates are always found (Dargnat et al. (2009) found phthalates concentrations around 2 µg/L). The metronidazole and norfloxacin were found at concentrations below 0.05 µg/L in the effluent (Clara et al., 2010).

Efficiency of organic pollutant removal in WWTP

Several studies have discussed the removal efficiency of wastewater treatment trains, that corresponds to the ratio of output-to-input OC concentration (Bolong et al., 2009; Gros et al., 2010; Fatta-Kassinos et al., 2011a; Jelic et al., 2011; Gao et al., 2012). Removal efficiency depends on OC themselves, treatment train and seasonal variations in OC concentrations (Vieno et al., 2005; Takao et al., 2008), as well as climatic conditions that affect for example water fluxes in the WWTP. Runoff dramatically reduces the OC removal rate: in a period of increased influent flow, the removal rate dropped to below 5% from over 60% previously (Ternes, 1998). The variability of removal efficiency can be pointed out also by comparing published results for the same OC: removal efficiency varies from 17% (Rosal et al., 2010) to 98% (Peng et al., 2006) for sulfamethoxazole, from 12% (Spongberg and Witter, 2008) to 80% (Karthikeyan and Meyer, 2006) for tetracycline, from 4.3% to 72% (Rosal et al., 2010) for erythromycin.

Two main mechanisms may be involved in the OC removal: sorption on sludge (or on other specific sorbents) and biodegradation (Carballa et al., 2004). Sorption on sewage sludge is the main removal process for hydrophobic OC having high sorption capacities, e.g. for several pharmaceuticals: fluoroquinolones (Golet et al., 2003) and tetracyclines (Kim et al., 2005). By contrast, sorption is negligible for most polar pharmaceuticals, the main possible removal process being then biodegradation, e.g. for acetaminophen, caffeine, salbutamol and salicylic acid whose degradation efficiency can exceed 90% in WWTP (Gomez et al., 2007; Jones et al., 2007). Others pharmaceuticals are hardly removed as gemfibrozil and fenofibric (Bendz et al., 2005), the ß-blockers acebutolol and sotalol (Vieno et al., 2006), the fluoroquinolones ciprofloxacin and norfloxacin (Lindberg et al., 2006; Vieno et al., 2006), and the iodinated X-ray contrast media iomeprol and iopromide (Ternes et al., 2007). Very low removals (<40%) were also reported for carbamazepine, diatrizoate, iopamidol and roxithromycin (Bendz et al., 2005; Bernhard et al., 2006; Vieno et al., 2006; Ternes et al., 2007). Radjenovic et al. (2009) found also that antiepileptic carbamazepine and diuretic hydrochlorothiazide are recalcitrant to biodegradation in the WWTP. The low degradation (<30%) of carbamazepine in WWTP was confirmed by Miao et al. (2005). Lincomycin was also another pharmaceutical little degraded (17%) (Karthikeyan and Meyer, 2006). Deblonde et al. (2011) found removal rates ranging from 0% (contrast media) to 97% for caffeine (psycho-stimulant) that may be considered as an indicator of human origin, whose concentration in influent is often the highest among the OC investigated.

Additional tertiary treatments may allow improving OC removal efficiency. Yang et al. (2011) followed the concentration of 19 pharmaceutically active compounds and personal care products in primary effluent (i.e. after pre-treatment) and after each step of the following tertiary treatment train: membrane filtration – adsorption on granular activated carbon - ozone oxidation. Caffeine and acetaminophen were found at the highest concentrations in the primary effluent (~10^5 ng/L), followed by ibuprofen (~10^4 ng/L), sulfamethoxazole and a DEET (~10^3 ng/L), and other compounds (concentrations on the order of several hundred of ng/L). After activated sludge treatment and membrane filtration, the concentrations of caffeine, acetaminophen, ibuprofen, phthalate, tetracycline, and 17α-ethynylestradiol had decreased by more than 90%. Erythromycin and carbamazepine, which were resistant to biological treatment, were eliminated by 74 and 88%, on average, by adsorption on granular activated carbon. Ozonation may oxidize most of the remaining compounds by >60%, except primidone and phthalate. Of the initial 16 compounds detected in the primary effluent, only sulfamethoxazole, primidone, caffeine and DEET were frequently detected in the final effluent, but at concentrations about 10–100 ng/L. The incomplete removal in the sewage treatment plants can be related to the low concentration of each compound, possibly not enough in relation to the catabolic enzyme affinities of sewage microbiota (Daughton and Ternes, 1999). Thus, reduction of concentration by dilution with fresh water can reduce the efficacy of biological treatment, suggesting treating specific pollutions at the source, rather than in WWTP collecting all wastewaters (Joss et al., 2006). Unfortunately, some OC have a chemical structure resisting to conventional wastewater treatments (phenols, chlorinated hydrocarbons, some pesticides …).

Among the personal care products, **fragrances (musks) are ubiquitous contaminants, persistent and bioaccumulative**, that are sometimes highly toxics (amino musk transformation products are toxicologically significant) (Daughton and Ternes, 1999). Musks are refractory to biodegradation that explains why there are difficult to remove in the wastewater treatment plants. Because synthetic musks are ubiquitous, used in large quantities, introduced into the environment almost exclusively via treated sewage effluent, and are persistent and bioconcentrated, they are indicators for the presence of other personal care products (Rimkus et al., 1997; Gatermann et al., 1998; Daughton and Ternes, 1999).

A general problem concerns the elimination of polar OC as acidic pharmaceuticals, but also other acidic compounds like some pesticides and benzotriazoles **which belong to the list of high production volume chemicals**. Benzotriazoles are largely used as corrosion inhibitors in many industrial applications, but also in households (dishwashing agents); their elimination efficiency in WWTP ranged between 20 to 70% (Weiss et al., 2006) and they are regularly discharged with municipal wastewater (Reemtsma et al., 2010). Among pesticides, azoles used as fungicides are often found in the wastewater, with a limited efficiency of the WWTP (between 30 and 65% of elimination) (Stamatis et al., 2010). **The dilution may be the only mechanism to reduce the concentration of some polar OC that are neither biodegradable nor retained by any of the natural or technical barriers** in the WWTP. A typical and well documented example of such a compound is ethylenediaminotetraacetate (EDTA), a chelating agent used in industrial processes as well as in consumer products. EDTA is not biodegradable under environmental conditions and may only be destructed by photolysis of its iron-complex in a natural environment. The effluents of 8 municipal WWTP in Western Europe were analysed over 10 months by liquid-chromatography−mass spectrometry for the occurrence of 36 polar pollutants, including household and industrial chemicals, pharmaceuticals, and personal care products. In a long-term study of the effluents of three WWTP. Reemtsma et al. (2006) showed that polar OC, sulfophenylcarboxylates and EDTA were detected above 10 µg/L on average, while benzotriazoles, benzothiazole-2-sulfonate, diclofenac, and carbamazepine showed mean concentrations of 1−10 µg/L, followed by some fire retardants, naphthalene disulfonates, and personal care products in the range of 0.1−1 µg/L. Half of the determined polar OC were not significantly removed in tertiary wastewater treatment (Reemtsma et al., 2006).

For some OC, the practices linked to the treatment of wastewater are the main sources of these OC in the environment, especially for some chlorinated compound produced during chlorine disinfection.

The degradability of a given OC can be described by laboratory experiments allowing the calculation of the rate constants of degradation (k_{bio}) considering that kinetic of pseudo first order. Using this approach, Joss et al. (2006) proposed the identification of three groups of OC according to their degradation constant: compounds with k_{bio}<0.1 $L.g^{-1}.d^{-1}$ are not removed to a significant extent (<20%), compounds with k_{bio}>10 $L.g^{-1}.d^{-1}$ are greatly transformed (>90%) and in-between moderate removal is expected. Briefly, only 4 targets compounds (ibuprofen, paracetamol, 17ß-estradiol and estrone) of the 35 compounds studied are degraded by more than 90%, while 17 compounds (including macrolides and sulfonamides) were removed by less than 50% during biological wastewater treatment (Joss et al., 2006).

Organic pollutant fate in the environment after wastewater spreading

Several studies investigated the occurrence and distribution of pharmaceuticals in soil irrigated with reclaimed water (Kinney et al., 2006; Ternes et al., 2007; Gielen et al., 2009).

Sorption on soils is an important process conditioning the OC fate in soils, as it regulates leaching. Because of the large spectra of molecular properties, **sorption properties may greatly vary between compounds** of a group of OC: e.g. the compilation of antibiotics sorption coefficients (K_d) showed that they vary between 0.2 and 6000 L/kg, which correspond to very mobile and immobile compounds, respectively. In general, **sulfonamide antibiotics and organophosphate biocides are mobile** in the environment, whereas **tetracycline, macrolide and fluoroquinolone antibiotics exhibit low mobility** (Boxall, 2008). In a study by Chefetz et al. (2008), the sorption-desorption behaviour and the mobility of carbamazepine, naproxen and diclofenac were studied in soil layers sampled from a plot irrigated with both freshwater and wastewater. Carbamazepine and diclofenac were significantly retarded in the 0-5 cm soil layer rich in soil organic matter (OM). Carbamazepine was not affected by the water type, whereas diclofenac exhibited a higher retardation factor in the freshwater leaching system. Naproxen exhibited significantly lower retardation factors than diclofenac but with a similar trend. In the 5-15 cm soil sample containing low OM, naproxen was highly mobile while carbamazepine and diclofenac were still retarded. In the 15-25 cm sample, all compounds exhibited their lowest retardation factors. Sorption data suggested that OM governs the studied OC interactions with the soil samples. **Both the quantity and the physicochemical nature of soil OM affect sorption interactions** (Chefetz et al., 2008). This study suggests that carbamazepine and diclofenac can be classified as slow mobile compounds in OM-rich soil layers. When these compounds pass this layer and/or are introduced into OM-poor soils, their mobility increases significantly. This emphasizes the potential transport of pharmaceuticals to groundwater due to intensive irrigation with reclaimed wastewater in OM-poor soils (Chefetz et al., 2008).

OC degradation in soil is another important process conditioning the capacity of soils to eliminate OC. However, persistent, non-degradable OC can be accumulated in soils, as for example phthalate esters which have accumulated in agricultural soils irrigated by wastewater in China (Zeng et al., 2008): phthalates were detected in all soil samples with the concentrations ranging from 0.195 to 33.6 µg/g and mainly originate from wastewater irrigation and sewage sludge application. Excessive accumulation of such compounds in agricultural soils may not only result in environmental contamination, but leads also to elevated phthalates uptake by crops, which may affect food quality. Other results found that **most antibiotics are not biodegradable under aerobic conditions** (Thiele-Bruhn, 2003; Alexy et al., 2004; Gartiser et al., 2007). **Photolysis can be an additional degradation process** depending on the modalities of wastewater irrigation. Some pharmaceuticals are particularly sensitive to the photolysis as this is the case of tetracyclines, fluoroquinolones, sulphanilic acid, tylosin, nitrofuran antibiotics … (Kümmerer, 2009). But most of the works on photodegradation have been performed in water: photochemical decomposition could play an important role in surface water as an additional elimination pathway or for effluent treatment (Viola et al., 2004; Edhlund et al., 2006; Hu and Coats, 2007), but the extrapolation to potential photodegradation at the soil surface must be done carefully.

Risk due to the organic contaminants in irrigation wastewaters

Contamination of conventional water resources

Although soil contamination by persistent OC is the first risk to be considered, **the OC accumulated in soils can be transported by leaching and runoff from soil surface into the groundwater and surface waters** after rainfall events (Pedersen et al., 2005; Topp et al. 2008), all the more that the use of sewage sludge as organic amendment or fertilizer in agricultural fields (Oppel et al., 2004) may be another source of pollution.

However **soil acts as a reactor allowing degradation and retardation to leaching of OC**. Little data are available on the persistence and effects in the environment. Although the continuous introduction of OC into the environment through continuous irrigation or other types of discharge practices can make them 'pseudo-persistent' (Fatta-Kassino et al., 2011b), analyses have shown that **most organic compounds are rapidly decomposed** is soils after irrigation (Focazio et al., 2008; Duran-Alvarez et al., 2009). Durand-Alvarez et al. (2009) showed that despite the continuous application of contaminants with wastewater over many years, the concentrations of acidic pharmaceuticals and endocrine disrupting compounds were generally lower than those expected considering the input amounts by a single irrigation event. Only **carbamazepine showed evidence of persistence in the soils** (Fatta-Kassinos et al., 2011b). Kinney et al. (2006) found that measurable but low concentrations of pharmaceuticals can be detected in soil irrigated with reclaimed wastewater. The residues of polycyclic aromatic hydrocarbons, polychlorinated biphenyls, chlorinated benzenes and phenols were investigated in soil in a study undertaken by Al Nasir and Batarseh (2008) in a field irrigated with wastewater; the concentration levels of all targeted compounds (like naphthalene, acenaphthylene, acenapthene, fluorine, phnanthrene, anthracene, pyrene, chrysene, *o*-cresol, *p*-cresol, 2,4-dimethylphenol, 2,6-dichlorophenol, 2,3,5-trichlorophenol, 2,3,4,6-tetrachlorophenol, etc.) found in soil irrigated with wastewater were much higher than for the reference site, indicating a source of contamination due to irrigation with wastewater. The depuration role of soil through filtration has been evidenced by Xu et al. (2009). These authors showed that after 4 months of turf field irrigation with reclaimed municipal wastewater, no OC was detected in the leachate draining through the 89-cm profile. Ibuprofen, naproxen, triclosan, bisphenol A, clofibric acid, and estrone were detected in the surface to 30-cm soil profiles. The screenings of pollution risk identified the same 6 compounds as having the potential to contaminate groundwater, and under conditions of turf grass irrigation, clofibric acid and ibuprofen would be most prone to cause the pollution. An interesting lysimetric study was done by Ternes et al. (2007). Lysimeters located in an irrigated agricultural field irrigated with wastewater effluents were monitored with regard to the occurrence of 52 pharmaceuticals and 2 personal care products. No differences in pollution of the groundwater were found for soils with and without addition of digested sludge. Most of the selected OC were never detected in any of the lysimeter, although they were present in the treated wastewater used for irrigation. However, some OC were detected up to several µg/L (diatrizoate and iopamidol, the antiepileptic carbamazepine and the antibiotic sulfamethoxazole) while the acidic pharmaceuticals, musk fragrances, oestrogens and ß-blockers were likely sorbed or transformed while passing the top soil layer. **Anionic species of pharmaceuticals are mobile in the soils, showed little elimination, and led to risks of groundwater contamination, whereas most cationic or neutral compounds are efficiently retained on the soils** (Siemens et al., 2008). Scheytt et al. (2007) confirmed that clofibric acid is a compound highly mobile and persistent. Other pharmaceutical detected after wastewater irrigation in the unsaturated zone were primidone and propyphenazone. In the column experiment in the lab no transformation and no retardation was found for clofibric acid, whereas diclefenac was degraded (79% of initial amount), and only 37% and 17% of degradation was found for ibuprofen and propyphenazone, respectively (Scheytt et al., 2007). Siemens et al. (2008) showed that pharmaceuticals found at high concentrations in the wastewater (>1 µg/L) had little elimination during soil passage pointing risk of groundwater contamination with naproxen, ibuprofen and diclofenac as a consequence of wastewater irrigation. In a monitoring study using wells close to a field irrigated with wastewater. Katz et al. (2009) noted low concentrations of carbamazepine in water indicating the persistence of this compound in the subsurface. Avisar et al. (2009) questioned the wastewater reuse practice for irrigation, because they point out the contamination of the aquifer under land irrigated with treated wastewater effluents for about 5 decades, they found out that concentrations of the OC in the groundwater were found to be up to 20 ng/L.

In addition to leaching, **runoff is another transfer process resulting into a risk of surface water contamination**. Topp et al. (2008) studied the runoff of pharmaceuticals and personal care products following application of sewage sludge to an agricultural field. Ibuprofen and acetaminophen concentrations in the runoff first decreased and then increased, suggesting that these OC were initially chemically or physically sequestered in the sewage sludge and subsequently released in the soil. Carbamazepine and triclosan were detected at low concentrations in a runoff event 266 d after sewage sludge application.

Uptake from soils and plants contamination by organic pollutants

Organic contaminants remaining in surface soils may be uptaken by plants. Very limited information is available in the literature. Previous research focused primarily on plant uptake of veterinary pharmaceuticals that are associated with animal wastes and demonstrated their potential to accumulate in plants, and some data are available on uptake of antibiotics from soil amended with manure containing antibiotics by carrot roots, lettuce leaves (Boxall et al., 2006) and corn (Kumar et al., 2005). The highest uptakes of sulfamethazine were found in corn and lettuce, followed by potato (Dolliver et al., 2007). The low concentration levels into the vegetal tissues allow deducing a very low health risk. The OC residues, which are reversibly adsorbed to soil, may be taken up by plants. In a greenhouse experiment, corn took up lasalocid and monensin. In laboratory experiments, it has been demonstrated the uptake of sulfadimethoxin in sorghum, pea and corn, and this uptake had an influence on their development (Schneider, 2008). Hydroponic culture plants, incorporated sulfanamide up to a final concentration of 180 to 2000 mg/kg. Roots of corn and sorghum accumulated much more active ingredients than the shoots. Similar results were obtained for rye, carrot, corn, sorghum and pea in field trials. Enrofloxacin was also accumulated in µg/g amounts (Schneider, 2008). In the case of a negative effect on plants as showed by Schneider (2008), it is not clear whether the effect resulted from the direct damage of the plant by OC or antibiotics effects on soil microorganisms was responsible for the damage by affecting the plant-microorganism symbiosis (Chander et al., 2005). The decay in the number of soil bacteria leads to a lack of feed for soil fauna (protozoa, nematodes, micro-arthropods) and finally influences soil functions: plant residues are decomposed slower, denitrification is slower, and therefore nutrients are recycled more slowly (Migliore et al., 1998). Al Nasir and Batarseh (2008) noted that different plants showed different uptakes of various OC from a soil irrigated with wastewater. Roots were the most contaminated part of the plant, while fruits were the least contaminated. The uptake ratios are dependent on the plant type and the physicochemical properties of organic compounds. In a greenhouse experiment, Wu et al. (2010) studied the uptake of three pharmaceuticals (carbamazepine, diphenhydramine, and fluoxetine) and two personal care products (triclosan and triclocarban) by soybean. Carbamazepine, triclosan, and triclocarban were found concentrated in root tissues and translocated into above ground parts including beans, whereas accumulation and translocation for diphenhydramine and fluoxetine was limited. **OC introduced by irrigation appeared to be more available for uptake and translocation than those introduced by sewage sludge**. The uptake from soil to root and translocation from root to leaf may be rate limited for triclosan and triclocarban and metabolism may occur within the plant for carbamazepine.

Ecotoxic effect of organic contaminants

Direct ecotoxic effects of OC present in wastewater used for irrigation are not treated in the literature. Ecotoxicology papers are focused on effects of wastewater on aquatic media. The reasons are that effluents of WWTP are usually connected to the water resources. As an example, Gros et al. (2010) showed that susceptibility for pharmaceuticals decreases in the order algae > daphnia > fish; they concluded that no significant risk could be associated to the presence of pharmaceuticals in effluents, mainly because of the dilution and low concentrations. Another example, coupling soil and water compartments is the study of Ternes et al. (2007) that found that potential estrogenic effects of wastewater disappeared after irrigation, since the most potent steroid oestrogens were not measurable. Only one paper proposed an original approach of integrated risk assessment using life cycle impact assessment methods (LCIA) (Muñoz et al., 2008). This method allows quantifying the potential environmental impacts on ecotoxicity and human toxicity of wastewater containing priority and emerging pollutants. This methodology was applied to wastewater influent and effluent samples from a WWTP. Characterization factors were applied to the average concentration of 98 OC, obtaining impact scores for different scenarios: discharging wastewater to aquatic recipient, and using it for crop irrigation. The results show that treated wastewater has a substantially lower environmental impact than the influent, and that pharmaceuticals and personal care products greatly contribute to toxicity in this wastewater. Ciprofloxacin, fluoxetine, and nicotine constitute the main personal care products of concern in this case study, while 2,3,7,8-TCDD, and hexachlorobenzene were the priority pollutants with highest contribution. When wastewater was released to fresh water ecosystems, the impact was mainly caused by fluoxetine, triclosan, and ciprofloxacin. Another scenario considered using wastewater for irrigation and thus releasing it to soil: the impact on terrestrial ecosystems is in this case mostly due to ciprofloxacin. Finally, the impact on human health of using wastewater for crop irrigation is mostly due to nicotine and gemfibrozil, attributing the impact to nicotine and hexachlorobenzene in the influent, and to 2,3,7,8-TCDD and hexachlorobenzene in the effluent (Muñoz et al., 2008).

Concluding remarks

Organic contaminants that may be present in raw wastewaters mainly include various compounds in personal care products, pharmaceuticals, phthalates, as well as some biocides used for material protection. However when there is a unique urban sewage network that collect both house wastewaters and runoff waters, they may also contain pesticides and polycyclic aromatic hydrocarbons (PAH). At least, some organic contaminants (solvents) may be produced into the WWTP during disinfection by chlorine. A classification has been proposed with the following 16 groups: steroids, non-prescription drugs, fragrances and flavors, antibiotics, pesticides, other prescription drugs, fire retardants, plasticizers, insect repellent, detergent metabolites, disinfectant, cosmetics, polycyclic aromatic hydrocarbons, solvents, dyes/resins/fuels and antioxidants. Concentrations of most OC in urban wastewaters (raw or treated) are generally below the toxic levels for humans, but potential problems may result from the combined effects of several pollutants and/or their cumulative consequences over long-term periods

The main conclusions concerning the content of pharmaceuticals in wastewaters are:

- Analgesics and anti-flammatories have been widely reported in sewage treatment effluents; the highest concentrations corresponding to the compounds sold in higher quantities: ibuprofen, diclofenac, acetaminophen, naproxen …;
- Antiobiotics: macrolides, sulfonamides, tetracycline, fluoroquinolones, chloramphenicol and trimethoprim have been identified in wastewater, in the inputs of the sewage treatment plants and in the sewage effluents. The most frequently detected antibiotic in environmental samples is the metabolite of the macrolide erythromycin (Hirsch et al., 1999). Although compounds of the penicillin class are used in the highest amounts, they have not generally been detected in the sewage effluents;
- ß-blockers: metoprolol, propranolol, betaxolol, bisoprolol, carazolol are frequently detected;
- Hormones and steroids: reproductive hormones (estradiol, estrone, hydroxyestrone) and contraceptive (ethinylestradiol) were generally detected at low concentrations in sewage effluents;
- Antidepressants: only fluoxetine was detected in sewage effluents;
- Antiepileptics: carbamazepine is the most often used antiepileptic and it and its metabolites have been frequently detected in sewage effluents at high concentrations.

Dealing with the removal efficiency in wastewater treatment plants and as a general trend of results in the literature, we can conclude some highlights:

- Psycho-stimulants seem to be easily removed, e.g. about 97% for caffeine (Ternes et al., 2007);
- The best then removed compounds in WWTP including an activated sludge systems are phthalates with removal efficiency above 90% (Bendz et al., 2005).
- The removal efficiency is about 71% for bisphenol A and about 50% for antibiotics (Gomez et al., 2007);

- Benzotriazoles removal is very variable, ranging between 20 to 70% (Weiss et al., 2006);
- Analgesics, anti-inflammatories and ß-blockers are resistant to treatments (removal efficiency of 30–40%) (Miège et al., 2009; Deblonde et al., 2011);
- Musks and derivatives are very persistent (Daughton and Ternes, 1999), as EDTA, metal-complexing agent (Reemtsma et al., 2006);
- Erythromycin and carbamazepine are resistant to biological treatment (Radjenovic et al., 2009; Yang et al., 2011).

Standards for OC have to be considered in regulations dealing with wastewater reuse for crop irrigation, the values being established from potential chemical risk assessments. It is particularly the case for the pharmaceutical products which are secreted after use by humans and arrive through the sewage system to the wastewater treatment plants (WWTP).