Salmonella in surface and drinking water: Occurrence and water-mediated transmission

Caterina Levantesi a,⁎, Lucia Bonadonna b, Rossella Briancesco b, Elisabeth Grohmann c, Simon Toze d, Valter Tandoi a

a Water Research Institute, CNR, via Salaria km 29, 300-00015 Monterotondo, Rome, Italy
b Department of Environment and Primary Prevention, Hygiene of Internal Water Unit, ISS, viale Regina Elena 299-00161, Rome, Italy
c Department of Infectious Diseases, University Medical Center Freiburg, Hugstetter Strasse 55, 79106 Freiburg, Germany
d CSIRO EcoSciences Precinct, Dutton Park 41 Boggo Road, Dutton Park, QLD 4102, Australia

Abstract

Salmonella is one of the leading causes of intestinal illness all over the world as well as the etiological agent of more severe systemic diseases such as typhoid and paratyphoid fevers. While water is known to be a common vehicle for the transmission of typhoidal Salmonella serovars, non-typhoidal salmonellae are mainly known as foodborne pathogens. This paper provides a brief review of the last ten years of peer reviewed publications on the prevalence of Salmonella in natural freshwaters and drinking waters, and on the relevance of these sources for Salmonella dissemination. In industrialized countries, Salmonella was rarely reported in water-borne outbreaks despite it being frequently detected in surface waters including recreational waters and waters used for irrigation or as a drinking water source. Consistent contamination with irrigation waters has been shown to be a common route of crop contamination in produces related Salmonella outbreaks. Multiple drug resistant (MDR) Salmonella strains, that represent an increased hazard for human health and that may contribute to the dissemination of drug resistances were also detected in surface water in developed countries. Surface runoff was shown to play a main role as driver of Salmonella load in surface waters. Accordingly, analysis of serovars indicated a mixed human and animal origin of Salmonella contribution to surface waters, emphasizing the role of wild life animals in water contamination. Data relating to Salmonella prevalence in surface and drinking water in developing countries are quite rare. Nevertheless, data on water-borne outbreaks as well as case control studies investigating the risk factors for endemic typhoid fever confirmed the relevance of water as source for the transmission of this disease. In addition epidemiological studies and Salmonella surveys, consistently provided an undeniable evidence of the relevance of MDR Salmonella Typhi strains in water-borne typhoid fever in developing countries.

© 2011 Elsevier Ltd. All rights reserved.
1. Introduction

One third of the world's population live in countries with some level of water stress and water scarcity is expected to increase in the next few years due to increases in human population, per capita consumption and the resulting impacts of human activity on the environment (Asano, Burton, Leverenz, Tsuchihashi, & Tchobanoglous, 2007). The availability of good quality water sources is therefore getting more and more limited and the impact of water-borne pathogens in human health is expected to be significant (Suresh & Smith, 2004). It is therefore critical to understand the relevance of natural and drinking water contribution to transmission of pathogenic microorganisms.

Salmonella is a ubiquitous enteric pathogen with a worldwide distribution that comprises a large number of serovars characterized by different host specificity and distribution. This microorganism is one of the leading causes of intestinal illness through the world as well as the etiological agent of more severe systemic diseases such as typhoid and paratyphoid fever (Pond, 2005). Zoonotic salmonellae are commonly described as foodborne pathogens, however, drinking water as well as natural waters are known to be an important source for the transmission of these enteric microorganisms (Ashbolt, 2004; Leclerc, Schwartzbrod, & Dei-Cas, 2002). Salmonella, just like other enteric bacteria, is spread by the fecal-oral route of contamination. This microorganism can enter the aquatic environment directly with feces of infected humans or animals or indirectly, e.g., via sewage discharge or agricultural land run off. Overall Salmonella spp. and subspecies can be found in a large variety of vertebrates. Beside humans, animal sources of Salmonella include pets, farm animals and wild animals; calves, poultry, pigs, sheep as well as wild birds (sea gull, pigeon) and reptiles can all be reservoirs of Salmonella (Dolejská, Bierosová, Kohoutová, Literák, & Cízek, 2009; Lightfoot, 2004; Wray & Wray, 2000). Plants, insects and algae were also found capable of harboring Salmonella and might be implicated in the transmission of this enteric pathogen (Byappanahalli et al., 2009; Guo, van Iersel, Garcia-Migura, Perez-Piñeiro, & Saco, 2004; Polo et al., 1999; Wilkes et al., 2009).

Salmonellae are frequently found in environmental samples. They are usually present in large numbers in raw sewage ($10^3$–$10^4$ CFU/L) and can still be present in wastewater effluent after advanced secondary treatment including coagulation, filtration and disinfection (Maier, Pepper, & Gerba, 2000; Wéry, Lhoutellier, Ducray, Delgenes, & Godon, 2008). Soil and sediment were also found to harbor salmonellae (Abdel-Monem & Dowidar, 1990; Gorski et al., 2011; Tobias & Heinemeyer, 1994) and sediment particles are believed to function as a micro ecological niche enhancing salmonellae survival in lakes (Chandran et al., 2011). In the aquatic environment this pathogen has been repeatedly detected in various types of natural waters such as rivers, lakes, coastal waters, estuarine as well as contaminated ground water (Haley, Cole, & Lipp, 2009; Levantesi et al., 2010; Martinez-Urtaza et al., 2004; Martinez-Urtaza, Liebana, Garcia-Miguera, Perez-Piñeiro, & Saco, 2004; Polo et al., 1999; Wilkes et al., 2009).

In addition to its widespread occurrence, elevated survival capacities in non-host environment have been reported for Salmonella (Winfield & Groisman, 2003). The growth of Salmonella in non-host environments such as wastewater sludge and compost has also been reported (Zaleski, Josephson, Gerba, & Pepper, 2005) and the growth of Salmonella in water supplies is also considered possible due to its ability to colonize surfaces and replicate in biofilms of distribution system pipes (Jones & Bradshaw, 1996). However, standard disinfection procedures used in drinking water treatment processes are active against salmonellae (Cicmanec, Smith, & Carr, 2004).

Taxonomically the genus Salmonella comprises two species namely S. hongori and S. enterica. The species S. enterica is further differentiated into six subspecies (enterica, salamae, arizonae, diarizonae, indica and houtenae) among which the S. enterica subspecies enterica is mainly associated to human and other warm blooded vertebrates. Traditionally members of the genus Salmonella are clustered in serovars according to their flagellar (h) and somatic (O) antigens. Currently over 2400 Salmonella serovars have been described but only about 50 serovars, all within the subspecies enterica, are common causes of infections in humans and warm blooded animals (Popoff, 2001). Salmonella serovars have different host specificity, diverse geographic distribution, and cause different syndromes. On the basis of the clinical syndromes caused Salmonella are divided in to two distinct groups namely the typhoidal and non-typhoidal Salmonella serovars (Pond, 2005).

Enteric fevers, typhoid and paratyphoid fever are severe, contagious systemic diseases caused by the infection of the serovars Typhi and Paratyphi. Even though not common in developed countries, enteric fevers remain an important and persistent health problem in less industrialized nations. Overall, in 2003 an annual incidence of approximately 17 million cases of typhoid and paratyphoid fevers was reported worldwide (Kindhauser, 2003). Differently from other Salmonella serovars, Typhi and Paratyphi are host adapted and can only infect humans; stools of infected persons are therefore the original source of contaminations for these pathogens. Water contaminated with feces of human cases and carriers is one of the main vehicles of typhoid fever infections.

Differently from typhoidal Salmonella strains, non-typhoidal salmonellae, the ubiquitous subtypes found in a number of animal species, are more frequently associated to foodborne than to water-borne transmission. These zoonotic Salmonella serovars tend to cause acute but usually self-limiting gastroenteritis. In some patients, however, these same serovars can cause severe systemic diseases such as osteomyelitis, pneumonia and meningitis (Pond, 2005). Severe and invasive presentations of non-typhoidal salmonellosis in Africa are common in children with co-morbidities (Graham, 2002) and immune compromised HIV infected adults (Gordon, 2008). Notably the emergence of MDR strains of Salmonella Enteriditis and Typhimurium was associated to the observed incidence of invasive salmonellosis (Gordon & Graham, 2008).

The increased frequency of MDR Salmonella strains in human infections is an emerging issue of major health concern (Lightfoot, 2004; Lynch et al., 2009; Pond, 2005). As a consequence, the possible role of fecally contaminated waters in the dissemination of MDR Salmonella, both typhoidal and non-typhoidal, as well as antibiotic resistance genes through horizontal gene transfer is of great interest. This issue is of particular concern in developing countries where the common use of untreated, low quality water as drinking water sources together with inappropriate use and self-prescribing of antibiotics increase the risk of drug resistance and the probability of widespread dissemination of resistant strains of high human health concern (Oluuye, Dada, & Odeyemi, 2009; Srikanthia et al., 2007).

Overall, the above reported characteristics suggest that water might be an important source for the transmission of all salmonellae, not only for the typhoid serovars. An increased knowledge on the prevalence, diversity and survival of this enteric bacterium in the environment, together with a proper assessment, trough effective
surveillance and epidemiological studies, of the relevance of the water environment in the transmission of Salmonella disease is desirable. In this respect the possible involvement of water in food contamination is also of great interest.

This paper provides a brief review of our understanding of the contribution of natural fresh water and drinking water as a route of Salmonella contamination. Initially epidemiological data on the water-mediated transmission of Salmonella infections and on the possible strategies to control these diseases are described. Successively recent literature relating to Salmonella prevalence, diversity, and survival in surface water and drinking water is summarized. Finally, a short overview of the most relevant emerging issues which the scientific community is actually addressing in relation to Salmonella in water environments is given. In particular the importance of water environment in the transmission of drug resistant Salmonella strains and the possible role of water in fresh food contamination are described. Furthermore the use and the evaluation of alternative fecal indicators of water contamination for Salmonella monitoring in aquatic environment is reported. Considering that limited scientific publications exist that report the presence of Salmonella in water when compared to reports of Salmonella in food, peer reviewed scientific publications of the last ten years were reviewed.

2. Water-borne Salmonella infections and control strategies

Two different types of data are the most common available in respect to Salmonella water-mediated infections: the description of water-borne salmonella outbreaks (Berg, 2008; Bhunia et al., 2009; Farooqui, Khan, & Kazmi, 2008; Franklin et al., 2009; Kozlca, Claudet, Solomon, Dunn, & Carpenter, 2010; Swaiddiwudhipong & Kanlayanaphotporn, 2001; Taylor, Sloan, Cooper, Morton, & Hunter, 2000) and the analysis of risk factors associated with endemic or sporadic Salmonella infections (Dennho et al., 2009; Gasem, Dolmans, Keuter, & Djokomelojanto, 2001; Sharma, Ramakrishnan, Huttin, Manickam, & Gup, 2009; Srikanthiah et al., 2007; Tran et al., 2005; Vollaard et al., 2004). Beside scientific publications, reports of national surveillance systems, such as Communicable Disease Surveillance Centre (CDSC, United Kingdom) and Centers for Disease Control and Prevention and Environmental Protection Agency (CDCEPA USA), provide useful data on salmonellae water-borne disease outbreaks (Crain et al., 2010; Dale, Kirk, Sinclair, Hall, & Leder, 2007; Smith et al., 2006). The level and quality of water-borne disease surveillance data, however, varies in different countries. In particular the situation in developing countries is problematic due to the lack of laboratory facilities and the scarcity of economic resources, however surveillance systems in many European countries have also been judged incapable of detecting water-borne disease (Blasi, Carere, Pompa, Rizzuto, & Funari, 2008; Hunter, 2003; Stanwell-Smith, Andersson, & Levy, 2003).

2.1. Developing countries

Literature data related to water-borne salmonellae in developing countries relate mostly the typhoid Salmonella serovars. In the less industrialized area of the world, in particular in the Indian sub-continent and South East Asia, typhoid and paratyphoid fevers occur both in epidemic and endemic form, and remain a major public health problem (DeRoeck, Jodar, & Clements, 2007). The burden of typhoid fever worldwide is further compounded by the spread of multiple drug resistant S. Typhi (Kim, 2010; Lynch et al., 2009; Srikanthiah et al., 2006; Swaiddiwudhipong & Kanlayanaphotporn, 2001). Most of the recent publications on typhoid and paratyphoid fever water-borne infections in developing countries are from the Asian continent (Bhunia et al., 2009; Farooqui et al., 2009; Gasem et al., 2001; Kim et al., 2003; Lewis et al., 2005; Luxemburger et al., 2001; Mermin, Villar, & Carpenter, 1999; Sharma et al., 2009; Srikanthiah et al., 2007; Swaiddiwudhipong & Kanlayanaphotporn, 2001; Tran et al., 2005; Vollaard et al., 2004). Very few or no publication from Africa and Central/Latin America, respectively, addressed the role of water as vehicle of Salmonella transmission in the last ten years.

Reports on typhoid and paratyphoid fever epidemics in Asia consistently indicate that contamination of drinking water, sourced from well water (Farooqui et al., 2009), piped municipal drinking water (Bhunia et al., 2009; Kim et al., 2003; Lewis et al., 2005; Mermin et al., 1999) and unboiled spring water (Swaiddiwudhipong & Kanlayanaphotporn, 2001) was the main source of outbreaks. In addition to this, most of the papers (Kim, 2010; Lewis et al., 2005; Mermin et al., 1999; Swaiddiwudhipong & Kanlayanaphotporn, 2001) clearly show the involvement of MDR typhoid strains in water-borne epidemics. Mermin et al. (1999) reported a massive epidemic of MDR typhoid fever in Dushanbe, Tajikistan, associated with the consumption of municipal water. In the first half of the year 1997, 8901 cases of typhoid fever and 95 associated deaths were reported in Dushanbe. A case-control study demonstrated that Salmonella Typhi infection was associated with drinking unboiled water or obtaining water from sources outside the house. Lack of chlorination, equipment failure, and back-siphonage in the water distribution system led to contamination of the drinking water (Mermin et al., 1999). Of 29 Salmonella serovar Typhi isolates tested, 27 were resistant to seven antibiotics, namely to ampicillin, chloramphenicol, nalidixic acid, streptomycin, sulfisoxazole, tetracycline, and trimethoprim-sulfamethoxazo. Al- though 93% of the tested Salmonella serovar Typhi isolates from Dushanbe were resistant to antimicrobials commonly used for typhoid fever, the case mortality rate of 1.0% was rather low (Mermin et al., 1999).

Farooqui et al. (2009) reported the study of a community outbreak of typhoid fever associated with drinking water in a village close to Karachi, Pakistan. This outbreak claimed three human lives and left more than 300 people infected within a one week. The infected people consumed water from a well that was the only available source of drinking water in the village. Epidemiological investigations revealed the gross contamination of the well with dead and decaying animal bodies, their fecal material and garbage. Microbiological testing confirmed the presence of MDR strains of Salmonella serovar Typhi in 100% of the well water samples and 65% of household water samples (Farooqui et al., 2009). In the summer of 2002 during a 7-week period, 5963 cases of typhoid fever were reported in Bharatpur, a town in Nepal with a population of 92,214 inhabitants (Lewis et al., 2005). This outbreak was the largest single-point source outbreak of MDR typhoid fever reported so far. The outbreak was traced with molecular epidemiological methods to a single source—the sole municipal water supply. A total of 90% of the isolates were resistant to more than one antibiotic. MDR S. Typhi were also found to be the etiological agent in a water-borne outbreak in a non-endemic community with otherwise good sanitation in Thailand (Swaiddiwudhipong & Kanlayanaphotporn, 2001).

In agreement with the studies relating to typhoid fever outbreaks, epidemiological studies on the risk factors for endemic and sporadic typhoid fevers in Asia confirmed the association between this disease and the use of poor quality water (Gasem et al., 2001; Sharma et al., 2009; Srikanthiah et al., 2007; Tran et al., 2005). Srikanthiah et al. (2007) and Tran et al. (2005) indicated the use of untreated drinking water in Uzbekistan and unboiled surface water in Vietnam, respectively, as a major risk factor for typhoid fever. Srikanthiah et al. (2007) also showed that MDR Salmonella Typhi strains were important in endemic typhoid fever in the Samarkand region of Uzbekistan. They found that multiple antimicrobial resistance to ampicillin, chloramphenicol, and trimethoprim-sulfamethoxazo, were in 6 (15%) of 41 Salmonella serovar Typhi isolates from patients. Furthermore recent consumption of antimicrobials (two weeks preceding illness) additionally increased the risk of infection (Srikanthiah et al., 2007). In Semarang city and the surrounding area (Indonesia), typhoid fever
was associated with living in a house without water supply from the municipal network along with the use of low quality water for washing and drinking (Gasem et al., 2001). Finally, Sharma et al. (2009) showed that the unsafe use of water, namely scooping out water from containers instead of using piped water or water stored in a narrow mounted container at home, was associated with typhoid fever. Using GIS to explore the epidemiological pattern across Vietnam by investigating the risk factors associated with shigellosis, typhoid, and cholera, Kelly-Hope et al. (2007) revealed that typhoid fever prevailed in the Mekong River Delta. They observed that typhoid fever was mostly associated with high vapor pressure and the use of river and streams as drinking water. On the contrary, forested regions and public tap drinking water were found to be negatively associated with typhoid fever. In contrast to this, Vollaard et al. (2004) showed that febrile contaminated drinking water was not associated with typhoid and paratyphoid fevers in Jakarta Indonesia. However, in this study, the habit of boiling water before consumption was declared by all the people surveyed, both test cases and control sources, possibly explaining the lack of association between microbiological contamination of drinking water and typhoid fever. Vollaard et al. (2004) also showed that typhoid fever was significantly associated with consuming ice cubes and ice drinks highlighting this as a likely water mediated route of transmission. The importance of ice cubes as a potential source of typhoid infection is supported by other studies in which it was shown that consumption of ice cubes (Gasem et al., 2001), flavored ice drinks (Black et al., 1985) and ice creams (Luby et al., 1998) are risk factors for this disease. In agreement with these observations was the report that prolonged survival of Salmonella in ice (Butler, Mahmoud, & Warren, 1977; Dickens, DuPont, & Johnson, 1985).

Nevertheless most of the recent risk factor studies showed that use of unsafe water was not the only risk factor associated with typhoid/paratyphoid fever thus indicating multiple routes of transmission for these diseases (Gasem et al., 2001; Sharma et al., 2009; Vollaard et al., 2004). Interestingly, Vollaard et al. (2004) showed that in Jakarta typhoid and paratyphoid fevers are associated with different routes of transmission. Risk factors for typhoid and paratyphoid fevers, respectively, were mainly within and outside the household. In other developing countries, typhoid, and paratyphoid fevers are the commonest cause of bacteraemia in children under 5 years (Gordon & Graham, 2008). Life-threatening invasive disease outbreaks in children caused by MDR non-typhoidal salmonellae have been reported in several African countries (e.g., Graham, 2002; Kariuki et al., 2006). Nevertheless the sources and mode of transmission of non-typhoidal salmonellae in Africa are still unknown. In a prospective study in Kenya, Kariuki et al. (2006) compared the serovars and genotypes of non-typhoidal salmonellae isolates from children with bacteraemia, from family contacts, and from home environmental samples. Salmonella isolates from family contacts showed the highest similarity to those of clinical cases indicating that human-to-human transmission route plays a major role in non-typhoidal salmonellae transmission in the area. Nevertheless, clustering of non-typhoidal Salmonella bacteraemia in the rainy season in Africa suggests that there is also a water-borne/water-associated transmission route (Suresh & Smith, 2004).

2.2. Developed countries

In contrast to what has been observed for the less industrialized nations, most of the data on water-borne salmonellae in developed countries involve non-typhoidal Salmonella serovars. In many industrialized countries, the widespread implementation of municipal water and sewage treatment systems in the second half of the 20th century has resulted in a dramatic decline in the incidence of water-borne typhoid fever (Lynch et al., 2009; Smith et al., 2006). Even if they remain endemic in a few geographic regions (Rizzo et al., 2008), typhoid and paratyphoid fever are now rare diseases, mainly associated with people returning from foreign travel to regions where these diseases are still much more common (Lynch et al., 2009).

In the USA since 1971, the CDC, in collaboration with the EPA and with the Council of State and Territorial Epidemiologists, began a surveillance program to collect and report data of water-borne outbreaks. According to the CDC data from 1971 to 2000, non-typhoidal zoonotic Salmonella were the causes of 15 drinking-water-borne outbreaks which accounted for 6% of the total zoonotic water-borne outbreaks in the USA (Craun, Calderon, & Craun, 2004). As revised by these authors, most of these salmonellae outbreaks (11/15) were associated with community water systems and groundwater instead of surface water. In the same period, the use of untreated groundwater along with inadequate treatment of collected groundwater and distribution systems contamination were the most important deficiencies identified for causing outbreaks of enteric bacteria. Further analysis has shown that various Salmonella subspecies, comprising Typhimurium, Enteriditis, Bareilly, Javiana, Newport and Weltevreden, were the causes of water associated Salmonella outbreaks in the USA (Craun et al., 2004). From 2000 to 2006 salmonellae were found very rarely or not at all in drinking water-borne outbreaks in USA (Blackburn et al., 2004; Liang et al., 2006; Yoder et al., 2008). Nevertheless, two recent outbreaks indicated that water-borne salmonellae are still of health concern in cases of deficiency of water treatment or inadequate water supply systems in the USA (Berg, 2008; Kozlca et al., 2010). In August 2008, the health department and regulatory officials identified an outbreak of...
Salmonella I4,[5],12:i− involving five persons in a rural community in Tennessee. An untreated water supply was recognized as the source of infection. It was found that the water, which had been collected from a spring, had been stored in a small unprotected reservoir subject to contamination from runoff and wildlife (Kozliva et al., 2010). In the second reported case during March and April 2008, an outbreak of water-borne disease associated with Salmonella struck the city of Alamosa in Colorado. Salmonellae were found to have contaminated the public water system that supplies drinking water to the community. The outbreak resulted in 442 reported illnesses, 122 of which were laboratory-confirmed, and one death. Epidemiological estimates suggested that up to 1300 people may have been ill (Berg, 2008). Alamosa’s drinking water comes from deep artesian wells in an aquifer considered to be a protected groundwater source. Prior to the outbreak, the city’s drinking water was not chlorinated for disinfection but was historically in compliance with all health-based drinking water standards. The most probable cause of the outbreak was a contamination of a ground-level water storage reservoir by animal feces that successively spread throughout the entire system. The reservoir was found to have several small cracks and holes that likely allowed the contamination source to enter.

Only one Salmonella water-borne outbreak related to recreational water exposure was recorded by the CDC between 1995 and 2005 (Levy, Bens, Craun, Calderon, & Herwaldt, 1998; Pond, 2005). On the contrary, recreational aquatic environments, both salt and fresh waters, were indicated as the most important risk factor for sporadic childhood Salmonella infections at three Washington state county health departments (Denno et al., 2009). Denno et al. (2009) conducted a prospective case control study to investigate the possible association between sporadic childhood reportable enteric infections and different plausible exposures. Infection with Salmonella was found to be strongly related to playing or swimming in natural water sources. In addition, associations between the use of private wells as home drinking water sources and the use of septic systems for home wastewater disposal were also observed as potential sources of Salmonella infections.

In Australia, salmonellae were found to be the most common pathogen in drinking water-borne outbreaks from 2001 to 2007 (Dale et al., 2007). Salmonella spp. were implicated in five of 10 observed drinking water-borne outbreaks and in an outbreak derived from a contaminated aquarium. The serovars Saintpaul, Typhimurium, subsp. I11b 61:i:i25, Muenchen and Para B by java, were identified as outbreak causes. Differently from what has been observed in the USA where Salmonella outbreaks were mostly related to contaminated community water systems, in Australia contaminated tank and bore waters were identified as the origin of outbreaks. Ashbolt and Kirk (2006) conducted a case control study to investigate risk factors for sporadic Salmonella Mississippi infections in Tasmania (Australia) showing that the exposure to untreated drinking water was a risk factor for this disease. These authors also highlighted that most of the exposures to untreated drinking water were recorded as exposure to water collected in rainwater collection tanks. Roof collected rain water were involved in several other water-borne outbreaks by Salmonella in Australia (Franklin et al., 2009; Taylor et al., 2000) suggesting that appropriate preventive measures must be undertaken to avoid that the increased use of rainwater tanks may increase the risk of water-borne disease outbreaks (Franklin et al., 2009).

In Europe, monographic water-borne outbreaks reports such as those of CDC/EPA, are not available because drinking water is defined as food and thus included in foodborne outbreaks reporting. Country specific review of water-borne outbreaks in Italy (Blasi et al., 2008) and the UK (Smith et al., 2006) were recently published. According to the complete surveillance data of Communicable Disease Surveillance Centre (CDSC), Salmonella was never associated to water-borne outbreaks in England and Wales from 1992 to 2003 (Smith et al., 2006). In contrast, salmonellae were implicated in outbreaks that occurred in Italy from 1998 to 2005. However because the surveillance system on communicable diseases in Italy is not planned to efficiently identify water-borne outbreaks, the number of reported cases do not represent an accurate clear estimation of the real situation (Blasi et al., 2008).

2.3. Control strategies to prevent Salmonella drinking water related transmission

As for other water-borne pathogens, different measures can be adopted to hinder the spread of Salmonella in water environment, and consequently reduce human risk of infection. Regarding the measures to reduce the occurrence of Salmonella in drinking water, the control strategies have to be elaborated and adopted on the basis of the water resource availability and the economic and developmental level of the country. For this purpose, the World Health Organization has developed Guidelines for Drinking Water Quality, which provides an internationally harmonized support to help countries develop rules and standards that are suitable to national and local circumstances (WHO, 2008).

In developing countries, commonly lacking piped water supply, self-sustaining decentralized approaches including point of use chemical and solar disinfection, safe water storage and behavioral changes are indicated as reliable options to directly target the most affected population and reduce water-borne disease burden through improved drinking water quality (Mintz, Bartram, Lohery, & Wegelin, 2001). In this respect, three recent reviews that used meta-analysis to evaluate the effect of water treatment at point of use on water quality and diarrhea reduction showed that point of use interventions more efficiently reduced diarrhea incidence, with respect to water source interventions (Arnold & Colford, 2007; Clasen, Schmidt, Rabie, Roberts, & Cairncross, 2007; Fewtrell et al., 2005).

Studies of the risk factors for typhoid and paratyphoid endemics and epidemics infections confirm that simple decentralized control strategies might be effective in the control of these diseases (Sharma et al., 2009; Srikantiah et al., 2007). According to the findings of Sharma et al. (2009) home chlorination of drinking water, the use of narrow-mounted containers for safe water storage, and drawing out water by tilting the container or using taps to avoid contamination may be effective practices to avoid spread of typhoid in west Bengal India. Farooqui et al. (2009) reported measures recently introduced by WHO that include solar disinfection, bleach addition, boiling, and use of low cost ceramic filters, which are well suited to prevent typhoid fever outbreaks due to the consumption of contaminated surface or drinking water.

Education regarding the risk of drinking untreated surface water as well as promoting the use of boiled drinking water carried from home, was indicated by Srikantiah et al. (2007) as practical intervention measures to reduce water related transmission of typhoid fever in Samarkand (Uzbekistan). Srikantiah et al. (2007) also suggested specific intervention strategies to reduce the increased risk linked to MDR typhoid strains. According to their results the use of antimicrobials two weeks before onset of typhoid fever was independently associated to this disease; hence the development of primary care clinical treatment guidelines to reduce unnecessary antimicrobial exposure may efficiently decrease the risk of typhoid fever.

In many industrialized countries, the success of applied control strategies is confirmed by the small number of water-borne outbreaks caused by salmonellae. Nevertheless, outbreaks caused by microbial contamination of drinking water still result in substantial human and economic costs in these countries (Berg, 2008; Risbore et al., 2007). The identification of the causes outbreaks and the potential threats to water quality is necessary to formulate effective prevention strategies that will minimize the further onset of outbreaks and related costs. Causes of water-borne outbreaks in developed countries have been extensively reviewed in recent publications (Craun et al., 2010; Craun,
outbreaks

3. Role of contaminated waters in Salmonella foodborne outbreaks

In recent years, reports of Salmonella outbreaks related to fresh produce consumption have illustrated the likely relationships between salmonellae, water and ultimately foodborne Salmonella infections (CDC, 2002, 2008; Greene et al., 2008; Mohle-Boetani et al., 2009; Sivapalasingam et al., 2003). It is now commonly accepted that fruit and vegetable consumption is a risk factor for infections with enteric pathogens (Heaton & Jones, 2007). Three recently published reviews clearly summarized the current knowledge on the role of fresh produce as source for the transmission of human enteric pathogens (Berger et al., 2010; Heaton & Jones, 2007), and in particular of Salmonella (Hanning, Nutt, & Ricke, 2009). Salmonella is the most common aetiologic agents associated to fresh produce related infections (Heaton & Jones, 2007; Sivapalasingam, Friedman, Cohen, & Tauxe, 2004). A wide spectrum of produce have been associated with salmonellae infections including tomatoes (Greene et al., 2008; Sivapalasingam et al., 2004), serrano peppers (CDC, 2008), cantaloupe (CDC, 2002; Sivapalasingam et al., 2004), lettuce (Takkinen et al., 2005), basil (Pezzoli et al., 2008), and mango (Sivapalasingam et al., 2003). Salmonellae are often isolated from produce samples in routine surveys (Thunberg, Tran, Bennett, & Matthews, 2002). Additionally some S. enterica strains have the ability to adhere to plant surfaces where they are able to survive for long periods and then grow (Gandhi, Golding, Yaron, & Matthews, 2001; Islam et al., 2004). The capacity of this pathogen to become endophytic, thus to invade internal plant parts has also been reported (e.g. Deering, Pruitt, Mauer, & Reuhs, 2011; Klers, Franz, van Gent-Pelzer, Zijlstra, & van Bruggen, 2007; Lapidot & Yaron, 2009; Schikora, Carreri, Charpentier, & Hirt, 2008). Among others Lapidot and Yaron (2009) demonstrated the ability of S. enterica serovar Typhimurium to transfer from contaminated irrigation water in plant roots and then into the edible parts of mature parsley. However the transfer of S. enterica serovar Typhimurium from water to plants was shown only to occur with highly contaminated irrigation water (Lapidot & Yaron, 2009).

Water is likely to be an important source of produce contamination in the field as well as in post harvested processing (Berger et al., 2010). Water sources of varying microbiological quality are used for irrigation of produce worldwide. For example, in the UK, 71% of irrigation water is obtained from surface waters which receive treated sewage effluent (Tyrrel, Knox, & Weatherhead, 2006) and in developing countries untreated wastewater is also commonly used to irrigate crops, thus increasing the risk of microbial contamination (Ashbolt, 2004). Gagliardi, Milliner, Lester, and Ingram (2003) showed an increased count of microbial indicators on cantaloupe rinds after fruit washing and cooling in hydrocooler indicating that inadequately decontaminated water used in post harvesting produce processing can also be a sources of microbial contamination.

Contaminated irrigation and processing waters were indicated as possible sources of Salmonella contamination in several fresh produce outbreaks (CDC, 2002, 2008; Greene et al., 2008; Sivapalasingam et al., 2003). In 2002 and 2005 two multistate outbreaks of S. Newport in USA, caused by the same rare S. Newport strain, were associated with eating tomatoes (Greene et al., 2008). Tomatoes causing of the outbreak were traced back to the eastern shore of Virginia where the S. Newport outbreak strain was isolated. Notably this outbreak strain was isolated from the pond water used to irrigate tomato fields both in 2002 and 2005 (Greene et al., 2008). The presence of the S. Newport outbreak strain, two years apart, in the irrigation pond demonstrate that if not properly addressed, the source of contamination can persist and cause continuous outbreaks (Greene et al., 2008).

Salmonella serovar Newport was also the etiological agent in a multistate outbreak linked to mango consumption in USA (Sivapalasingam et al., 2003). The implicated mangoes were traced back to a single Brazilian farm where mangoes were washed with contaminated water (Sivapalasingam et al., 2003). Analysis of the irrigation and processing water at this farm indicated the presence of Salmonella spp. in the water. Sivapalasingam et al. (2003) suggested that hot and cool water treatment which allowed the internalization of Salmonella in the mangoes was the most likely source of contamination. The S. Newport outbreak strain was no actually isolated from the Brazilian water samples however, the farm investigation took place many months after the outbreak had occurred (Sivapalasingam et al., 2003). In 2008, a large multistate outbreak of Salmonella Saintpaul associated with multiple raw produce items infected a total of 1442 people in the USA and Canada (CDC, 2008). Jalapeno and serrano peppers imported from Mexico were identified as the major sources of this outbreak. The S. Saintpaul outbreak strain was isolated from samples of jalapeno pepper as well as of irrigation water at a Mexican farm suggesting a possible route of Salmonella contamination (CDC, 2008). Irrigation of fields with sewage contaminated waters and processing produce with Salmonella contaminated waters were also indicated as possible sources of contamination in an outbreak of S. Poona infections in the USA associated with eating imported cantaloupe (CDC, 2002).
Several studies have investigated the potential source of produce contamination in the supply chain both at the pre-harvested and post-harvested stages. According to the epidemiological data, surveys of *Salmonella* contamination in water used for fresh produce irrigation and in food processing identified the possible involvement of these waters in fresh vegetable and fruit contamination (Castillo et al., 2004; Duffy et al., 2005; Espinoza-Medina et al., 2006; López Cuebas, León Félix, Jiménez Edeza, & Chaidez Quiroz, 2009). *Salmonella* was detected in irrigation waters used for different type of fresh produce items, such as tomatoes, peppers and cantaloupes (Castillo et al., 2004; Duffy et al., 2005; Espinoza-Medina et al., 2006; López Cuebas et al., 2009). Espinoza-Medina et al. (2006) evaluated the incidence of *Salmonella* contamination during cantaloupe production and processing, in five farms in Sonora (Mexico) showing that irrigation water (23% of positive samples) and hands of workers packing cantaloupe (16.7% of positive samples) were the most likely sources of *Salmonella* contamination. Similarly, Castillo et al. (2004) described an extensive survey of *Salmonella* contamination at cantaloupe production farms in South Texas (USA) and Colima State (Mexico). By repetitive sequence based PCR analysis Castillo et al. (2004) identified the irrigation water source as a possible source of cantaloupe contamination. Similar levels of *Salmonella* contamination were found in the cantaloupe from USA and from Mexico (Castillo et al., 2004). Duffy et al. (2005) analyzed samples from fresh produce (orange, parsley and cantaloupe) and environmental samples (water, soil, equipment) at a produce production farm in Texas. *Salmonella* was only isolated from the irrigation water (16/25 isolates), equipment (6/25 isolates) and washed cantaloupe (3/25 isolates). However, by PFGE the *Salmonella* strains isolated from the irrigation water and equipment were shown to be different from the cantaloupe isolates (Duffy et al., 2005). Finally López Cuebas et al. (2009) recovered *Salmonella* in 39% of irrigation water samples in four different regions of the Cúillacan valley (Sinaloa Mexico), an important area for the production of crops exported in the USA. Notably, resistance to tetracyclines was observed in many of the *Salmonella* strains isolated which correlated with the use of these antibiotics to treat and control plant pathogens infections (López Cuebas et al., 2009; Lugo-Melchor et al., 2010).

4. *Salmonella* prevalence, diversity in surface, drinking water and groundwater

*Salmonella* contaminated waters might contribute through direct ingestion of the water or via indirect contamination of fresh food to the transmission of this microorganism. *Salmonella* prevalence in surface water and drinking water has not been uniformly investigated in different countries in recent papers. Surveys of *Salmonella* in fresh surface water environment were mainly performed in industrialized nations, in particular in Canada and North America. Reports of *Salmonella* prevalence in drinking water were instead more frequently from developing nations reflecting the higher concern relating to the use of low quality drinking water in these countries. Overall, the scientific community has mainly recently focused on the prevalence of this microorganism in impacted and non-impacted watersheds (Haley et al., 2009; Jokinen et al., 2011; Patchanee, Molla, White, Line, & Gebreyes, 2010), on the identification of the routes of salmonellae contamination (Gorski et al., 2011; Jokinen et al., 2010, 2011; Obi et al., 2004; Patchanee et al., 2010), and on the influence of environmental factors on the spread of *Salmonella* in water (Haley et al., 2009; Jokinen et al., 2010; Meinersmann et al., 2008; Wilkes et al., 2009). Particular attention was also addressed to the evaluation of available and new approaches to monitor sources of *Salmonella* contamination, and to predict its presence in different aquatic environments (Savichtcheva, Okayama, & Okabe, 2007; Schriewer et al., 2010; Walters, Cannon, & Field, 2007; Wilkes et al., 2009). Additionally, the recently available data on the prevalence of antibiotic resistant *Salmonella* strains in surface and drinking water (Bhatta et al., 2007; Dolejska et al., 2009; Meinersmann et al., 2008; Oluyeye et al., 2009; Patchanee et al., 2010) consistently indicate that these aquatic environments are a reservoir of MDR strains, which might contribute to the dissemination of these MDR strains through susceptible populations.

4.1. Surface waters

A survey of recent studies showed an increasing interest over the last two years on the role of non-host habitats, such as surface water environments as natural reservoir and in the transmission of *Salmonella* and other enteric pathogens (Ahmed, Sawant, Huygens, Goonetilleke, & Gardner, 2009; Byappanahalli et al., 2009; Dolejska et al., 2009; Haley et al., 2009; Jokinen et al., 2010, 2011; Patchanee et al., 2010; Schriewer et al., 2010; Wilkes et al., 2009). A summary of the available data on *Salmonella* prevalence in surface fresh waters sources is shown in Table 1.

The data provided in Table 1 confirms the ubiquitous nature of this enteric pathogen. Salmonellae were detected in different countries and in very diverse water sources, ranging from pristine (e.g. Patchanee et al., 2010; Till, McBride, Ball, Taylor, & Pyle, 2008) and low impacted water (e.g. Jokinen et al., 2010; Meinersmann et al., 2008; Patchanee et al., 2010) to heavily impacted water sources (e.g. Bonadonna, Filetici, Nusca, & Paradiso, 2006; Jyoti et al., 2010). *Salmonella* contamination occurred in surface water used for recreational purposes (Till et al., 2008); as source of drinking water (Till et al., 2008); and for irrigation (e.g. Gannon et al., 2004). This demonstrated wide distribution of salmonellae underlines that there is the potential for inadvertently using water contaminated with *Salmonella*, with associated potential health risks. As shown in Table 1, the *Salmonella* detection frequencies were extremely variable in the investigated surface waters with detection rates ranging from 3 to 100%. In general, the highest frequencies were reported in watersheds that had been highly impacted by human activities (Bonadonna et al., 2006; Jyoti et al., 2010) or in geographical areas with a history of high salmonellosis cases (Haley et al., 2009). However, Patchanee et al. (2010) showed that elevated frequencies of *Salmonella* were also found in a forestry watershed not influenced by human activities. Notably, these authors observed similar prevalence of salmonellae in watersheds with diverse potential contamination sources, namely residential industrial (58.8%), forestry (57.1%), crop/agriculture (50%), and swine production (41%).

Also of all the studies listed in Table 1, despite the importance of the knowledge of *Salmonella* concentrations in water to evaluate the risk of water related infection, only a few performed a quantitative assessment of *Salmonella* in environmental waters (Bonadonna et al., 2003, 2006; Byappanahalli et al., 2009; Haley et al., 2009; Jyoti et al., 2010; Lemarchand & Lebaron, 2003).

Temporal and spatial variation of *Salmonella* frequencies were commonly observed in surface water (Bonadonna et al., 2006; Byappanahalli et al., 2009; Haley et al., 2009; Jokinen et al., 2010; Lemarchand & Lebaron, 2003; Meinersmann et al., 2008; Till et al., 2008; Wilkes et al., 2009). It is possible that variations in the occurrence of *Salmonella* in ambient water may be governed, in part, by environmental parameters such as temperature, water chemistry, and solar radiation that influence survival and transport of the microorganism. Other possible reasons include changes in pathogen loading due to enhanced shedding from human and animal host, or from enhanced flow of water from contaminated sources. Positive correlations between rainfall events and *Salmonella* prevalence and/or diversity have been reported by many authors (Baudart, Lemarchand, Brisabois, & Lebaron, 2000; Haley et al., 2009; Jokinen et al., 2010; Polo et al., 1999; Schets, van Wijnen, Schijven, Schoon, & de RodaHusman, 2008; Wilkes et al., 2009). These results, which are supported by similar observations with respect to *Salmonella* contamination in
Table 1
Salmonella occurrence in surface waters.

<table>
<thead>
<tr>
<th>Level and frequency positive samples % (total samples)</th>
<th>Surface water</th>
<th>Dominant impact in the analyzed water</th>
<th>Country</th>
<th>Survey extent</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>8.5% (342)</td>
<td>River water</td>
<td>Mainly influenced by rainfall runoff and drainage from agricultural land</td>
<td>Canada</td>
<td>9 sampling sites, 2 years</td>
<td>Jokinen et al. (2011)</td>
</tr>
<tr>
<td>13% (186)</td>
<td>River water</td>
<td>Mixed</td>
<td>Canada</td>
<td>4 sampling sites, 2 years</td>
<td>Jokinen et al. (2010)</td>
</tr>
<tr>
<td>10³–10⁴ CFU/100 mL*</td>
<td>River water</td>
<td>Mixed comprising untreated sewage of residential setup</td>
<td>India</td>
<td>8 sampling sites, 1 day</td>
<td>Jyoti et al. (2010)</td>
</tr>
<tr>
<td>87% (8)</td>
<td>Four differently impacted watersheds systems</td>
<td>Swine production Residential/industrial</td>
<td>USA</td>
<td>4 sampling sites, 2 years</td>
<td>Patchanee et al. (2010)</td>
</tr>
<tr>
<td>7% (143)</td>
<td>River and estuaries</td>
<td>Not described</td>
<td>USA</td>
<td>10 sampling sites, 14 month</td>
<td>Schriewer et al. (2010)</td>
</tr>
<tr>
<td>3% (32)</td>
<td>Urban pond and tidal creeks</td>
<td>Mixed: WWTPs effluents, surface runoff, agricultural</td>
<td>Australia</td>
<td>8 sampling sites, 2 months</td>
<td>Ahmed et al. (2009)</td>
</tr>
<tr>
<td>0.002–0.017 MPN/mL</td>
<td>Lake and river water</td>
<td>Not described</td>
<td>USA</td>
<td>numerous sampling sites, 2 years</td>
<td>Byappanahalli et al. (2009)</td>
</tr>
<tr>
<td>16% (87)</td>
<td>Pond</td>
<td>Urban</td>
<td>Czech Republic</td>
<td>2 sampling sites, 1 year</td>
<td>Dolejská et al. (2009)</td>
</tr>
<tr>
<td>2.5–36.3 MPN/Lb</td>
<td>River watershed</td>
<td>Mixed: mainly forested and agricultural in an area with high incidence of salmonellosis mixed</td>
<td>USA</td>
<td>6 sampling sites, 12 months</td>
<td>Haley et al. (2009)</td>
</tr>
<tr>
<td>4–15% (1600)*</td>
<td>River and streams</td>
<td>Mixed: birds, dairy farming, municipal, sheep/pastoral, forestry undeveloped</td>
<td>Canada</td>
<td>24 sampling sites, 3 years</td>
<td>Wilkes et al. (2009)</td>
</tr>
<tr>
<td>75% (83)</td>
<td>Small river</td>
<td>Mainly rural area</td>
<td>USA</td>
<td>82 sampling sites, 1 day</td>
<td>Meinersmann et al. (2008)</td>
</tr>
<tr>
<td>3.8% (79)</td>
<td>Urban canals/river and recreational lakes*</td>
<td>Mixed comprising WWTP discharge</td>
<td>Netherlands</td>
<td>8 sampling sites, 1 year</td>
<td>Schets et al. (2008)</td>
</tr>
<tr>
<td>10.6% (216)</td>
<td>Stream water Puddles</td>
<td>Not described</td>
<td>Mexico</td>
<td>Random sampling, 2 years</td>
<td>Simental and Martinez-Urtaza (2008)</td>
</tr>
<tr>
<td>17.6% (17)</td>
<td>10% (725)*</td>
<td>Mixed: birds, dairy farming, municipal, sheep/pastoral, forestry undeveloped</td>
<td>New Zealand</td>
<td>25 sampling sites, 15 months</td>
<td>Till et al. (2008)</td>
</tr>
<tr>
<td>53% (30)</td>
<td>River and pond around Sapporo city</td>
<td>Mixed: WWTPs effluents, urban sewage, cow and pig farms</td>
<td>Japan</td>
<td>5 sampling sites, 3 months</td>
<td>Savichtcheva et al. (2007)</td>
</tr>
<tr>
<td>10–10⁶ MPN/100 mL</td>
<td>River†</td>
<td>Mixed: Urban/industrial comprising direct sewage discharge</td>
<td>Italy</td>
<td>8 sampling sites, 2 years</td>
<td>Bonadonna et al. (2006)</td>
</tr>
<tr>
<td>75% (375)*</td>
<td>River, borehole</td>
<td>Animal and human activities, human and animal feces</td>
<td>South Africa</td>
<td>8 sampling sites</td>
<td>Obi et al. (2004)</td>
</tr>
<tr>
<td>5.4% (802)</td>
<td>River and irrigation water</td>
<td>Mixed comprising WWTP discharge</td>
<td>Canada</td>
<td>16–21 sampling sites, 2 years</td>
<td>Gannon et al. (2004)</td>
</tr>
<tr>
<td>10²–10⁴ MPN/100 mL</td>
<td>River</td>
<td>Mixed: Urban/industrial, WWTP discharge agricultural</td>
<td>Italy</td>
<td>1 sampling site, 1 year</td>
<td>Bonadonna et al. (2003)</td>
</tr>
<tr>
<td>57% (14)</td>
<td>River water</td>
<td>Runoff from agricultural land and pastures, human and animal feces</td>
<td>Africa</td>
<td>Weekly sampling, 5 months</td>
<td>Obi, Potgieter, Bessong, and Matsaung (2003)</td>
</tr>
<tr>
<td>0.06–42.4 CFU/L</td>
<td>River</td>
<td>Mixed</td>
<td>France</td>
<td></td>
<td>Lemarchand and Lebaron (2003)</td>
</tr>
</tbody>
</table>

* qPCR quantification.
† Range of average monthly density.
‡ Different percentage in different seasons.
§ Only found in canals and river.
$ 15% of river sample used for drinking water supply.
$ Highly contaminated area at environmental risk.

Environmental samples including water and sediments (89%) and food (11%).

coastal area and marine environment (Baudart, Grabulos, Barrusseau, & Lebaron, 2000; Simental & Martinez-Urtaza, 2008), indicated that increased pathogen load could be associated with rainfall events as one major environmental driver of Salmonella contamination in surface water environment. In agreement with this a positive correlation was reported in the literature between rainfall events and the incidence of Salmonella water related infections (e.g. Haddock & Mallay, 1986; Suresh & Smith, 2004). In contrast, the variable seasonal occurrence of Salmonella in aquatic environments reported in the recent literature was not consistent with the summer peak of Salmonella observed in the fall (Wilkes et al., 2009) potentially reflecting enhanced livestock based fecal input during this period. According to Wilkes et al. (2009) salmonellae’s incidence was found to be positively correlated with tributary discharge values as well as with accumulative rainfall. Only Haley et al. (2009) in a study of a watershed in Georgia USA found that Salmonella incidence peaked in the summer period. Over the complete study period, the presence of this pathogen was positively correlated with water temperatures and rainfall events (1 or 2 days before sampling). According to these results, the authors concluded that multiple environmental factors, including enhanced pathogen loading during storm events, increased survival in the water environment at warm temperatures and increased host shedding in the warmer season might explain the Salmonella peak during the summer months.

Spatial differences in Salmonella prevalence in surface waters have also been reported in the recent literature (Bonadonna et al., 2006; Lemarchand & Lebaron, 2003; Meinersmann et al., 2008; Savichtcheva...
et al., 2007; Schriewer et al., 2010). Schriewer et al. (2010) reported a higher frequency of salmonellae in surface waters that have marine influences while Bonadonna et al. (2006) and Lemarchand and Lebaron (2003), observed that the highest concentration of this pathogen was associated to sampling points in densely populated areas and/or downstream of sewage effluents discharge sites.

MDR strains have been detected in surface water in developed countries (Dolejská et al., 2009; Meinersmann et al., 2008; Patchanee et al., 2010). In particular, Patchanee et al. (2010) showed that swine production was a major source of antibiotic resistant Salmonella in water environments as all the Salmonella isolates from a swine production watershed had multiple resistances to antibiotics. In contrast, antibiotic resistant strains were not detected from watersheds impacted by forestry, crop agriculture, or a residential/industrial area (Patchanee et al., 2010). Antibiotic resistant Salmonella have also been detected in surface water from a pond in the north-eastern part of the Czech Republic as well as in birds, black-headed gulls nesting in this pond (Dolejská et al., 2009). A total of 16% of water and 24% of gull samples yielded Salmonella. Antibiotic resistance was found in 12% of salmonellae isolated from the water and 28% of gull salmonellae. Resistance to the quinolone nalidixic acid, streptomycin, and tetracycline was often detected in these Salmonella isolates. Dolejská et al. (2009) concluded from these results that black-headed gulls can be important reservoirs of antibiotic resistant salmonellae. Meinersmann and co-workers investigated the prevalence of Salmonella and their antimicrobial resistance profile in a synoptic study of the Oconee River Basin, a small river in north-eastern Georgia, USA (Meinersmann et al., 2008). Water samples were obtained from 83 sites with Salmonella isolated from 62 of the samples, however, only seven of the isolates were resistant to any antimicrobials.

4.2. Diversity of Salmonella strains isolated from surface waters

All Salmonella serovars are considered as potential pathogens. Only about 50 of these serovars, however, have been predominantly isolated from humans or animals (Popoff, 2001). Information regarding the distribution and characteristics of Salmonella serovars present in the environment are essential in assessing the role of non-host aquatic habitats in the diffusion of Salmonella subtypes of high human health concern. Furthermore, the identification of source-specific serovars might help future tracking of sources of Salmonella contamination events.

The first comprehensive studies describing the diversity of Salmonella serovars in the aquatic environment were presented by Polo et al. (1999) and Baudart, Lemarchand, et al. (2000). Both research groups analyzed a large number of isolates from different natural aquatic systems (river, wastewater and marine coastal areas) showing a great diversity of Salmonella serovars. Since then, several works have provided further data on the different serovars present and on the possible sources of Salmonella contamination in surface water. A summary of recent available data is reported in Table 2.

With the exception of few studies that analyzed a limited number of isolates (Dolejská et al., 2009; Schets et al., 2008) or monitored highly contaminated watersheds (Bonadonna et al., 2006), the total number of serovars detected in the investigated aquatic environments ranged from 10 to 20 independently of the number of isolates or samples analyzed in the different studies. In comparison to the results of the initial studies by Baudart, Lemarchand, et al. (2000) and Polo et al. (1999), these latter consistent figures found that there was a low diversity of Salmonella serovars in the non-host water environment (Table 2). This difference might be explained by the very long period (5 years) of Salmonella monitoring described by Polo et al. (1999) and the reported increase in the numbers of serovars during flood events (10 of the 35 serovars were only isolated during the flood by Baudart, Lemarchand, et al. (2000)).

<table>
<thead>
<tr>
<th>Serovar number/total isolates</th>
<th>Diversity</th>
<th>Common serovars (percentage of total isolates)</th>
<th>Others serovars</th>
<th>Country</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>11/29</td>
<td>Rubilaw (72.4%)&lt;sup&gt;a&lt;/sup&gt;, t:11r:4&lt;sup&gt;a&lt;/sup&gt;, Derby</td>
<td>Give&lt;sup&gt;b&lt;/sup&gt;, Heidelberg&lt;sup&gt;b&lt;/sup&gt;, Typhimurium, Senftenberg, Mbandaka, Other</td>
<td>Canada</td>
<td>Jokinen et al. (2011)</td>
<td></td>
</tr>
<tr>
<td>12/104</td>
<td>Anatum (18.3%)&lt;sup&gt;c&lt;/sup&gt;, Gaminara (18.3%)&lt;sup&gt;c&lt;/sup&gt;, Inverness (18.3%)&lt;sup&gt;c&lt;/sup&gt;, Muenchen (8.7%)&lt;sup&gt;b&lt;/sup&gt;, Newport (8.7%)&lt;sup&gt;b&lt;/sup&gt;, Bredeney (6.7%)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>Mbandaka, Bullbay, Give, Miami, Braenderup</td>
<td>USA</td>
<td>Patchanee et al. (2010)</td>
<td></td>
</tr>
<tr>
<td>13/197</td>
<td>S. enterica subsp arizonae (40.6%)&lt;sup&gt;b&lt;/sup&gt;, Muenchen (14.2%)&lt;sup&gt;c&lt;/sup&gt;, Rubilaw (13.2%)&lt;sup&gt;c&lt;/sup&gt;, Mlikwasiina (6.1%)&lt;sup&gt;c&lt;/sup&gt;, Montevideo&lt;sup&gt;d&lt;/sup&gt;</td>
<td>Braenderup, Anatum, I47:z4z32, Gaminara, Liverpool, Bareilly, Saint Paul, J4(5):b</td>
<td>USA</td>
<td>Haley et al. (2009)</td>
<td></td>
</tr>
<tr>
<td>6/17</td>
<td>Enteritidis (50%)&lt;sup&gt;e,f&lt;/sup&gt;</td>
<td>Infantis, Derby&lt;sup&gt;e&lt;/sup&gt;, Indiana, Hadar&lt;sup&gt;e&lt;/sup&gt;, Typhimurium</td>
<td>Czech Republic</td>
<td>Dolejská et al. (2009)</td>
<td></td>
</tr>
<tr>
<td>19/73</td>
<td>Muenchen (20%), Rubilaw</td>
<td>Hartford, Give</td>
<td>USA</td>
<td>Meinersmann et al. (2008)</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>Newport Virchow</td>
<td>Typhimurium</td>
<td>Netherlands</td>
<td>Schets et al. (2008)</td>
<td></td>
</tr>
<tr>
<td>11/23</td>
<td>Urbana Suberu Vejle</td>
<td>Typhimurium (39%), Otmarschem, Soerenga, augustenberg, Breda, Coelo, Dju</td>
<td>Mexico</td>
<td>Simental and Martinez-Urtaza (2008)</td>
<td></td>
</tr>
<tr>
<td>45/92&lt;sup&gt;e&lt;/sup&gt;</td>
<td>Typhimurium (13.7%), Subsp. II (8.4%), Infantis (5.3%), Bredeney (4.2%), Stanley (4.2%), Derby (4.2%)</td>
<td>Saint Paul, Bovismorificants, Montevideo, Enteritidis, Give (only serotypes detected also by other authors)</td>
<td>Italy</td>
<td>Bonadonna et al. (2006)</td>
<td></td>
</tr>
<tr>
<td>19/36</td>
<td>Derby (16.6%), 03,10:r:4&lt;sup&gt;e&lt;/sup&gt;, Anatum</td>
<td>03,10:r:4&lt;sup&gt;e&lt;/sup&gt;, Subsp. IIIb, Subsp. IV, Stanley, Typhimurium, Tennessere, London, Bovismorificants, Dessau, Paratyphi B, Schleissheim, Lexington, Mbandaka, Enteritidis</td>
<td>Vietnam</td>
<td>Phan et al. (2003)</td>
<td></td>
</tr>
<tr>
<td>35/413&lt;sup&gt;b&lt;/sup&gt;</td>
<td>Typhimurium (31.1%), Virchow Panama Newport</td>
<td>Hadar Grunnepensis</td>
<td>France</td>
<td>Baudart, Lemarchand, et al. (2000)</td>
<td></td>
</tr>
</tbody>
</table>

(continued on next page)
According to the dominance of Salmonella Paratyphi B, although very rare, has been detected in Salmonella main sources of surface water contamination by enteric bacteria such as livestock was described by Patchanee et al. (2010). Patchanee et al. Salmonella (57.1% of samples) from a forestry watershed not impacted it was neither detected in the analyzed sewage (human contamination source) in the Little River rural watershed (Georgia, USA), Haley et al., 2009. This indicates that the Salmonella population in water environments is influenced by the incidence of human infections in the local area. Clinical serovars were not always dominant isolates in the studies of aquatic environment, however (Haley et al., 2009; Jokinen et al., 2011; Patchanee et al., 2010) suggesting that human sewage was not the sole or main source of contamination in these habitats. In agreement with this, the spectra of Salmonella serovars observed by various authors in surface water showed a mixed human/animal origin (Dolejská et al., 2009; Haley et al., 2009; Jokinen et al., 2011; Patchanee et al., 2010).

One relevant fact that can be noted is that the clinical serovars that were common or dominant in the studied geographic areas, were usually also detected in the surface water samples (Bonadonna et al., 2006). This indicates that the Salmonella population in water environments is influenced by the incidence of human infections in the local area. Clinical serovars were not always dominant isolates in the studies of aquatic environment, however (Haley et al., 2009; Jokinen et al., 2011; Patchanee et al., 2010) suggesting that human sewage was not the sole or main source of contamination in these habitats. In agreement with this, the spectra of Salmonella serovars observed by various authors in surface water showed a mixed human/animal origin (Dolejská et al., 2009; Haley et al., 2009; Jokinen et al., 2011; Patchanee et al., 2010).

The common assumption that livestock production is one of the main sources of surface water contamination by enteric bacteria such as Salmonella has not been fully confirmed by recent literature data. According to the dominance of the S. enterica subspecies arizonae (40% of isolates) in the Little River rural watershed (Georgia, USA), Haley et al. (2009) suggested that local raptor population may be a significant source of the total Salmonella in this surface water. Jokinen et al. (2011) could only detect the serovar Rubislaw in the Oldman river watershed (Alberta, Canada) and in the feces of wild birds, while it was neither detected in the analyzed sewage (human contamination source) nor in feces of domestic animals. High frequency of Salmonella (57.1% of samples) from a forestry watershed not impacted by livestock was described by Patchanee et al. (2010). Patchanee et al. (2010) also showed that Salmonella serovar Gaminara, was dominant in this same water system. According to Gaertner, Hahn, Rose, and Forstner (2008) S. Gaminara is mostly associated to wild animals. As previously noted, black-headed gulls were identified as an important reservoir of antibiotic resistant Salmonella contamination in a pond in Czech Republic (Dolejská et al., 2009). All of these results indicate that wild animals can be one of the major sources of Salmonella contamination in the environment. In agreement with this, wild birds and other wild life have been identified as source of Salmonella water-borne outbreaks in different countries (Angulo et al., 1997; Schuster et al., 2005; Taylor et al., 2000).

Understanding the relative contribution of different possible contamination routes on Salmonella introduction in surface waters is a necessary precursor for the selection of specific control strategies. An increase of knowledge on how the Salmonella genotypes and phenotypes are related to different potential contamination sources is essential. In this context, valuable information has been generated by the comprehensive work of Jokinen et al. (2011), Patchanee et al. (2010), and Gorski et al. (2011) that can be used for the suggestion of appropriate control strategies in impacted catchments and water sheds. Jokinen et al. (2011) compared the serovars detected in the Oldman river watershed with those present in sewage (human source) and in animal feces as potential source of Salmonella contamination in the investigated area. Only four serovars isolated from water (Rubislaw, Heidelberg, Give and I:11r:-) were also detected in feces or sewage samples. However, most of these serovars spanned a broad host range and thus it was not possible to identify the single contamination sources. Salmonella Rubislaw, the most common serovar in the water samples, was only detected in wild bird feces, but, since a limited number of fecal samples of wild birds was analyzed, it was not possible to understand the real contribution of these birds on the water contamination with Salmonella. Nevertheless, serovars comparison in feces and water samples indicated that other animals found in the watershed were not contributing to the Salmonella contamination in the Oldmann river watershed (Jokinen et al., 2010).

Patchanee et al. used pulsed-field gel electrophoresis PFGE genotyping coupled with serotyping techniques for tracking Salmonella contamination in four watersheds in North Carolina (USA). Salmonella genotypic and phenotypic diversity was analyzed and compared in differently impacted watersheds: swine-production, residential/industrial, forestry and crop agriculture (Patchanee et al., 2010). The resulting Salmonella genotypic and phenotypic diversity in differently impacted watersheds: swine-production, residential/industrial, forestry and crop agriculture was compared showing that isolates from the swine-production watershed were distinctly different from other isolates both in their genotypes and serovars. This suggested that there was a specific source of contamination for this watershed. In particular the serovars Anatum and Bredney were only found in this watershed. The authors also showed that Salmonella strains isolated from residential/industrial and forestry were genotypically related and suggested that there was a potential cross contamination between these watersheds.

Gorski et al. (2011) investigated the incidence and diversity of salmonellae in a major produce region of California. As part of the study they also analyzed serovars and genotypes (PFGE) of Salmonella strains isolated from water, cattle and wildlife feces, soil/sediment and preharvested lettuce/spinach. The serovars detected in the water samples, S. Infantis, S. Give, S. Typhimurium and the monophasic types 6,8:d:- and 6.8::e.n.z15. were also isolated from wildlife feces. According to PFGE patterns, however, all the strains isolated from wildlife were distinguishable from those isolated from water. Nevertheless, some near matches indicated that crow and coyotes were potential sources for water contamination. These authors conclude that source tracking may be possible in these environments;
however, they stressed that the use of genotyping methods with higher resolution than PGFE would be necessary to determine the relatedness of very clonal serovars.

4.3. Drinking water and groundwater

A summary of recent available publications on Salmonella occurrence in groundwater and drinking water, from a combination of water either treated or untreated prior to use as public drinking water untreated is provided in Table 3. As shown in Table 3, Salmonella was detected with varying frequencies in the analyzed water. Several studies describe Salmonella prevalence in drinking water in African countries, mainly South Africa and Nigeria (Akinwunmi et al., 2006; Adewale et al., 2006; Momba et al., 2006; Oguntoke et al., 2009; Oluyege et al., 2009; Potgieter et al., 2005). Momba et al. (2006) reported poor microbiological quality of drinking water in a rural area of the Eastern Cape Province (South Africa) where Salmonella arizonae was detected in 100% of municipal drinking water samples. Similarly in the Limpopo Province (South Africa), samples water collected from standpipes of boreholes and from surface water used as drinking water source were always found to be positive for Salmonella (Potgieter et al., 2005). As shown in Table 3 lower frequencies of Salmonella in drinking water samples were reported from Nigeria compared to South Africa (Akinwunmi et al., 2006; Oguntoke et al., 2009; Oluyege et al., 2009) (Table 3). Nevertheless, Nigeria surface and groundwater used as drinking water sources in rural communities were also shown to be reservoirs of Salmonella (Akinwunmi et al., 2006; Oguntoke et al., 2009; Oluyege et al., 2009). In addition, the surveys of Salmonella prevalence in drinking water in Nigeria undertaken by Oluyege et al. (2009) showed that all the Salmonella strains isolated were resistant to several antibiotics. Furthermore, MDR resistance to more than 4 more than 4 antibiotics was observed in most of these isolates (Oluyege et al., 2009). It should be noted, however, that in contrast to what was observed in surface waters studies from other countries, most of these African publications relied on very few data points to link the water as reservoirs of salmonellae. A notable comparison is the comprehensive study of Bhatta et al. (2007), which provided undeniable evidence of the poor microbiological quality of the public drinking water supplied in urban Nepal. Bhatta et al. (2007) clearly showed the importance of the human host-specific strains Typhi and Paratyphi A, indicating the impact of human source contamination on the overall Salmonella input in the analyzed water source. Furthermore, they highlighted the common occurrence of MDR Salmonella strains which increased the risk connected to the use of these water supply systems. The literature demonstrate that a lack of hygienic conditions, non-appropriate water supply structures, and improper disinfection were consistently identified as the main cause of water contamination in developing countries (Akinwunmi et al., 2006; Bhatta et al., 2007; Momba et al., 2006; Oluyege et al., 2009).

Groundwater is becoming increasingly relied upon as a major source of water and the security of water quality for groundwater in developing nations is a major issue. Contamination of groundwater by microbial pathogens has been documented also in developed countries due to failures in well head protection, inadequate off-set and diffuse contamination sources (Bockelmann et al., 2009; Borchardt, Haas, & Hunt, 2004; Goss & Richards, 2008). Groundwater can be contaminated by a wide range of pathogens, but like for most water sources, enteric pathogens remain the greatest risk. Despite the common assumptions that all enteric pathogens are a contamination risk for groundwater (e.g. John & Rose, 2005) the majority of studies of pathogens in groundwater has focused on the presence of indicator microorganisms (e.g. Emmanuel, Pierre, & Perrodin, 2009; Wall, Pang, Sinton, & Close, 2008), risks posed by enteric viruses (e.g. Borchardt et al., 2004; Locas, Barthe, Margolin, & Payment, 2008), or protozoa (e.g. Boyer, Kuczynska, & Fayer, 2009). In contrast, the information on the presence of Salmonella spp. in groundwater is limited. In one of the few studies reported on Salmonella in groundwater, Li et al. (2009) determined that the consumption of groundwater was an independent risk factor for a number of confirmed cases of Salmonella choleraesuis in Taiwan. In another study of groundwater quality in Turkey, Ozler and Aydin (2008) detected Salmonella in 15% of the groundwater samples tested.

There is also information about Salmonella detection and behavior in groundwater from a study where recycled water has been intentionally recharged to aquifers during Managed Aquifer Recharge (MAR). Levantesi et al. (2010) were able to occasionally detect Salmonella in two aquifers used as MAR sites receiving recycled water, however, they detected Salmonella much less frequently than other pathogens such as Cryptosporidium and Giardia.

In Australia, where the issue of water scarcity has resulted in the promotion of the use of roof harvested rain water for drinking or other purposes, several studies have been undertaken investigating the microbiological quality of the collected rain waters. Ahmed et al. (2006) detected Salmonella in tanks collecting roof harvested rain water for indoor and outdoor use and indicated wild birds or animal fecs as likely source of contaminations. A risk assessment study using these results indicated that there was a risk of infection for Salmonella ingestion that exceeded the threshold value of 1 extra infection for 10,000 persons a year suggesting that roof harvested rainwater should be disinfected if used for potable water purposes (Ahmed et al., 2010). Another study also linked roof harvested rain water with Salmonella water-borne infection in Australia (Ashbolt & Kirk, 2006; Franklin et al., 2009) further highlighting the risks from Salmonella in roof harvested rain water.

### Table 3

<table>
<thead>
<tr>
<th>Level and frequency % positive sample (total samples)</th>
<th>Water source</th>
<th>Country</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>6 - 10^3 - 3.8 - 10^5 gc/L 10.7% (214) 50% (4)</td>
<td>Roof harvested rainwater a</td>
<td>Australia</td>
<td>Ahmed et al. (2010)</td>
</tr>
<tr>
<td>10^4 - 10^5 CFU/100 mL b</td>
<td>Drinking water supply</td>
<td>India</td>
<td>Jott et al. (2010)</td>
</tr>
<tr>
<td>23.6%</td>
<td>Well and borehole water c</td>
<td>Nigeria</td>
<td>Oguntoket et al. (2009)</td>
</tr>
<tr>
<td>13.3% d</td>
<td>Surface water e</td>
<td>Nigeria</td>
<td>Oluyege et al. (2009)</td>
</tr>
<tr>
<td>14% (300)</td>
<td>Tap water of urban water supply system</td>
<td>Nepal</td>
<td>Bhatta et al. (2007)</td>
</tr>
<tr>
<td>100% (8) f</td>
<td>Borehole standpipes water and surface water g</td>
<td>South Africa</td>
<td>Momba et al. (2006)</td>
</tr>
<tr>
<td>16% (18) f</td>
<td>Well and tap water h</td>
<td>Nigeria</td>
<td>Akinwunmi et al. (2006)</td>
</tr>
<tr>
<td>100%</td>
<td>Borehole standpipes water and surface water i</td>
<td>South Africa</td>
<td>Potgieter et al. (2005)</td>
</tr>
</tbody>
</table>

**gc:** gene copies.

a 35% for indoor use including drinking, showering and kitchen use.

b Presumptive Salmonella identification.

c Used untreated for drinking purpose.

d Percentage of the Gram negative isolates from water samples.

e Determined by qPCR but reported as CFU/100 mL.

f % of positive sampling sites.

5. Salmonella survival and monitoring in water environment

5.1. Salmonella survival

The majority of research on Salmonella survival in aquatic environment has been done in the last decades of the 20th century. Overall, these older studies investigated the physicochemical and biological factors influencing Salmonella survival in various water environments.
environments and compared the survival capacity of different enteric bacteria (e.g. Salmonella and E. coli).

In the last ten years this topic has been less frequently addressed. Chandran and Hatha (2005) described a microcosm study that investigated the relative survival of E. coli and Salmonella Typhimurium in estuary waters focusing on the effect of biological factors, dissolved organic and inorganic substances, and sunlight on the inactivation of these bacteria. The results showed that sunlight was the most important factor for inactivation, followed by biotic factors. The biotic factors were determined to comprise of a combination of predation by protozoa and bacteria, bacteriophage lysis, and competition with autotrophic microbes. They also found that dissolved organic substances present in the estuarine water, in the absence of the native biota, promoted survival and growth of these enteric bacteria. In contrast to what has been previously described in other studies (Mezrioui, Baleux, & Trousselier, 1995; Rhodes & Karton, 1988; Winfield & Groisman, 2003), E. coli cells showed a better survival capacity when compared to S. Typhimurium under all tested conditions. In another microcosm study by the same research group, an enhanced survival capacity of Salmonella in lake sediment with respect to the overlaying water was observed (Chandran et al., 2011). According to these results, the lake sediment offered some sort of protection for this enteric pathogen and, thus, potentially act as a reservoir of Salmonella.

The effect of sunlight on the persistence of S. enterica, E. coli and Campylobacter jejuni in river and seawater was also investigated by Sinton, Hall, and Braithwaite (2007). They showed that inactivation rates in sunlight were clearly higher than in the dark and that the extent of inactivation was directly related to the amount of insolation. Furthermore, in agreement to Chandran and Hatha (2005) they showed that S. enterica was inactivated up to 1.6 times faster than E. coli.

Pathogens have been documented to decay when introduced into aquifers and this information can be used for Managed Aquifer Recharge sites (Gordon & Toze, 2003; Toze, 2004). As with the detection of pathogens in groundwater, most information available focuses on the survival of enteric viruses (Naser, Glozman, & Nitzan, 2002; Toze, Hanna, Smith, Edmonds, & McCrow, 2004) and there is limited information specifically on the survival of Salmonella. Despite this, inactivation of Salmonella in groundwater has been noted in one study of pathogen decay at a Managed Aquifer Recharge site. Toze, Bekele, Page, Sidhu, and Shackleton (2010) observed that Salmonella had a 1 log reduction time in groundwater of 1 day. This is similar to decay times observed in groundwater for other bacteria such as coliforms (Gordon & Toze, 2003) but less than observed for enteric viruses or protozoa (Toze et al., 2010).

Recent studies indicate that secondary habitat may provide the required conditions to promote survival and replication of enteric microorganisms including Salmonella. In particular, Ishii et al. (2006) and Byappanahalli et al. (2009) showed that the filamentous green alga Chladowora in Michigan lake, could harbor salmonellae at much higher concentrations compared to the numbers in the surrounding water. According to these observations and to previous results showing that this alga can support the growth of enteric bacteria by providing nutrients in the algal excretions (Byappanahalli, Shively, Nevers, Sadowsky, & Whitman, 2003), Byappanahalli et al. (2009) hypothesize that Salmonella is likely to multiply on Chladowora in the water column.

5.2. Conventional and alternative microbial indicators for Salmonella monitoring in aquatic environment

The limits and advantages of conventional and alternative indicators of fecal contamination for pathogen monitoring in aquatic environment have been recently extensively reviewed (Field & Samadpour, 2007; Savichtcheva & Okabe, 2006). The efficiency of fecal indicator bacteria (FIB) to predict the presence/absence of Salmonella in aquatic environment remains a matter of debate in recent papers (Ahmed et al., 2009; Savichtcheva et al., 2007; Schriewer et al., 2010; Wilkes et al., 2009). Despite this, in a comprehensive study describing the occurrence of enteric pathogens in a Canadian watershed and their relationships with environmental parameters and FIB, Wilkes et al. (2009) showed that a good prediction of Salmonella presence can be obtained by FIB determination. In particular, they indicated that E. coli and fecal coliforms were the most appropriate indicators of Salmonella presence compared to Clostridium perfringens, enterococci and total coliforms. The identified E. coli threshold level that gave the greater probability of pathogen detection was > E. coli 89 CFU/100 mL. This E. coli threshold was determined to correlate with the identification of 89% of Salmonella positive samples. Schriewer et al. (2010) also observed that at a specific threshold level fecal coliforms as well as enterococci and total coliforms were good predictors of Salmonella presence in Californian rivers with sea intrusion. They also found, however, that these microbial indicators did not efficiently predict Salmonella in rivers that were not influenced by sea intrusion. In contrast with these results, no correlation were observed between FIB and Salmonella presence/absence in ponds and creeks in the Brisbane city area in Australia (Ahmed et al., 2009), or in the Small River basin in Georgia, USA (Meinersmann et al., 2008).

Another potential indicator are the Bacteroidales. Bacteroidales are fecal anaerobes that are present in feces in much higher densities than conventional FIB and have different host specificities (Field & Samadpour, 2007). Feces from different sources such as human, horse, pigs, dogs, and ruminant can be identified by Bacteroides host specific PCR primers which can allow source tracking of fecal contamination. Favorable results were obtained using this method in comparison to other source tracking approaches (Griffith, Weisburg, & McGee, 2003). Furthermore, due to their host specific distribution, these markers might predict the presence of certain pathogens associated to specific source of pollution.

The relationship between Bacteroides 16S rRNA genetic markers and Salmonella in aquatic environment has been addressed in a few papers (Savichtcheva et al., 2007; Schriewer et al., 2010; Walters et al., 2007). According to Savichtcheva et al. (2007), human and total Bacteroides 16S rRNA genetic markers in wastewater and river samples were positively correlated with the presence of Salmonella and showed a good predictive value for the presence of this pathogen. They found that the probability of its occurrence became significantly high (>70–80%) when the concentration of human Bacteroides genetic markers exceeded 105 copies/100 mL. No correlations were found with cow and pig Bacteroides 16S rRNA genetic markers in the same study. Differently from the results from Savichtcheva et al. (2007), two recent studies analyzing river and estuarine water environments showed no predictive value for various Bacteroidales host specific markers in association with Salmonella detection (Schriewer et al., 2010; Walters et al., 2007). Nevertheless, Walters and colleagues showed that ruminant specific Bacteroidales markers were more frequently detected in Salmonella positive samples (36%) compared to human specific markers (2.6%) (Walters et al., 2007). They concluded that agricultural runoff were a major contribution to the Salmonella contamination in the analyzed watershed while contamination from human sources was only a minor contribution.

6. Conclusions

This review on the occurrence and risk of Salmonella in water has demonstrated that Salmonella can be present in a variety of aquatic environments and that contamination can come from a range of sources. Salmonella has been detected in sources as diverse as rivers, lakes, pond, groundwater and drinking water, while sources of the bacterium have been like to sources that include inputs from human, domestic animals and wild life.
Extensive surveys of *Salmonella* spp. occurrence in surface waters in industrialized countries confirmed the ubiquitous nature of this enteric pathogen. Moreover, the positive correlations between *Salmonella* frequencies and rainfall events observed in many of the studies indicated that surface runoff plays a main role as driver of *Salmonella* load in aquatic environments. Analysis of isolated serovars consistently showed a mixed human and animal origin of *Salmonella* in surface water environments. This emphasizing the role of wild life animals in water contamination along with the more accepted livestock and human sources. Overall, a great complexity was observed in the structure of *Salmonella* populations in surface water environments. Studies revealed large serovars diversity, both in water and in likely sources of contamination as well as spatial and temporal variation in the detection of *Salmonella* types and frequencies.

In spite of the recognized potential of microbial source tracking, these methods have not been extensively used to investigate the sources of *Salmonella* contamination in the surface water environments. Nevertheless, the results obtained to-date suggest that microbial source tracking by genotyping methods in these environments may be possible and may be extremely useful to identify related strains in complex environmental *Salmonella* populations and to highlight specific transmission routes.

*Salmonella* serovars of human health concern were shown to be widespread in natural fresh waters although more rare environmental serovars were also frequently detected. At present little is known about the pathogenicity of these rarer serovars isolated from the environment and further research is required to understand the role of these serovars in water-borne disease dissemination. Regardless, human infections have been reported for most of the serovars detected in surface water including those usually associated with wild animals. A number of studies in developed countries detected clinically relevant *Salmonella* serovars in water utilized for indoor uses, irrigation and recreational uses, and of further concern, also detected MDR *Salmonella* strains in natural freshwaters. Overall, these results proposed that there was the likelihood of surface water-mediated *Salmonella* infections. This conclusion, however, is not supported by epidemiological water-borne outbreaks data showing rare involvement of surface waters, both as drinking water sources and for recreational use, in *Salmonella* outbreaks. Nevertheless, exposure to recreational water was indicated as the most important risk factor for childhood *Salmonella* infections indicating that different transmission routes may be implicated in the transmission of sporadic and epidemic diseases.

*Salmonella* was not one of the leading causes of reported water-borne outbreaks in developed countries, being usually reported less frequently than other pathogens as agent of water-mediated epidemics (e.g. *Cryptosporidium, Giardia, E. coli* O157:H7; norovirus, *Shigella*, and *Campylobacter*). Detected *Salmonella* outbreaks were mostly linked to contaminated drinking waters. Different types of drinking water (municipal water system, well, tank water) were indicated as sources of outbreaks, however, all the outbreaks were consistently associated with the use of untreated or inadequately treated water. Studies have shown that *Salmonella* is efficiently removed by conventional disinfection procedures utilized in water treatment processes and thus is of little risk in adequately treated water sources. Despite this, the true incidence of *Salmonella* water-borne diseases might be greater than is reflected in the reported outbreaks statistics as not all water-borne disease outbreaks are recognized or reported, especially in countries with a low level of water-borne outbreaks surveillance. In addition, *Salmonella* contaminated waters used to irrigate and wash produce crops have been implicated in a large number of food-borne outbreaks demonstrating that additional indirect routes of transmissions should be considered to correctly evaluate the role of water in *Salmonella* diseases dissemination.

The data on water-borne outbreaks in developing countries are not complete as national outbreaks surveillance systems are usually limited and in spite of the likely large number of studies performed in the field, the data produced have rarely been published. The data available in the scientific literature related to water-borne transmission of *Salmonella* in developing countries are mainly case control studies that have analyzed the risk for endemic typhoid *Salmonella*. The scientific data, overall, confirms that water is a common source for the transmission of this disease, but also highlight that other risk factors are associated with typhoid fever indicating multiple routes of transmission. Reports of massive water-borne *Salmonella* outbreaks as well as epidemiological studies on endemic typhoid and paratyphoid fevers underlined the relevance of MDR *Salmonella* strains in these countries. The risk of MDR strains dissemination is further exacerbated in less industrialized countries through the customs of inappropriate use and self-prescribing antibiotics.

The efficiency of conventional and alternative microbial indicators of fecal contamination to monitor *Salmonella* in water environments is still a matter requiring further study due to the need for rapid, specific and reliable methods that can be used for the control of environmental water quality. Most of the studies that looked for a link between indicators and *Salmonella* found inconsistent results on the relationship between the occurrence of FIB and *Salmonella*. Additionally, no significant improvements were achieved by applying alternative indicators to the *Salmonella* presence in surface waters. Bacteroidales were found to perform equally or slightly better than FIB in a direct comparison of their capacity to predict *Salmonella* presence. However the relationship between Bacteroidales and *Salmonella* were found to not be constant in surface water as inconsistent results were obtained in different studies.

This review has shown that there are still limitations on the understanding of *Salmonella* in water and their potential sources. *Salmonella* remain a significant health risk in developing nations and improved understanding of the role of water in the dissemination of *Salmonella* in these countries and on how to manage the risks. Of particular concern is the rise in the presence of multi-drug resistance in *Salmonella* and the causes of this drug resistance.

References


C. Levantesi et al. / Food Research International 45 (2012) 587–602


Haddock, R. L., & Malilay, J. (1986). The possible role of rainfall in spreading
Hunter, P. R. (2003). Principles and components of surveillance systems. In P. R. Hunter,
Levandoski, C. Levantesi et al. / Food Research International 45 (2012) 587–602