FAO Fisheries and Aquaculture Report No. 937 FIPM/R937 (En)

ISSN 2070-6987

Report of the

FAO EXPERT WORKSHOP ON THE APPLICATION OF BIOSECURITY MEASURES TO CONTROL *SALMONELLA* CONTAMINATION IN SUSTAINABLE AQUACULTURE

Mangalore, India, 19–21 January 2010

Report of the

FAO EXPERT WORKSHOP ON THE APPLICATION OF BIOSECURITY MEASURES TO CONTROL *SALMONELLA* CONTAMINATION IN SUSTAINABLE AQUACULTURE

Mangalore, India, 19–21 January 2010

FOOD AND AGRICULTURE ORGANIZATION OF THE UNITED NATIONS
Rome, 2010

Copies of FAO publications can be requested from:
Sales and Marketing Group
Office of Knowledge Exchange, Research and Extension
Food and Agriculture Organization
of the United Nations
E-mail: publications-sales@fao.org
Fax: +39 06 57053360
Web site: www.fao.org/icatalog/inter-e.htm

ISBN 978-92-5-106553-2

PREPARATION OF THIS DOCUMENT

This document contains the report of the FAO Expert Workshop on the Application of Biosecurity Measures to Control *Salmonella* Contamination in Sustainable Aquaculture" held from 19 to 21 January 2010 and hosted by the Microbial Resources Center (MIRCEN), College of Fisheries, Mangalore, India. This Workshop was supported by FAO Multidonor Partnership Programme (FMPP) under Objective D.1: Support to national biosecurity initiatives and policies. The Workshop consisted of presentation of background papers by the experts and drafting of the report in three Working Groups chaired by Ronold Lee, Balakrish Nair and Brett Koonse. The plenary sessions to discuss the draft were chaired by Alan Reilly. FAO technical support for this Workshop was provided by Iddya Karunasagar and Lahsen Ababouch from the Products, Trade and Marketing (FIPM) Branch of Fisheries and Aquaculture Department.

FAO
FAO Expert Workshop on the Application of Biosecurity Measures to Control *Salmonella* Contamination in Sustainable Aquaculture, Mangalore, India, 19–21 January 2010.
FAO Fisheries and Aquaculture Report No 937. Rome, FAO. 2010. 39p.

ABSTRACT

This document contains the report of the FAO Expert Workshop on the "Application of Biosecurity Measures to Control *Salmonella* Contamination in Sustainable Aquaculture" held in Mangalore, India from 19 to 21 January 2010. The experts reviewed the current scientific evidence regarding the pathogen *Salmonella enterica*, its occurrence and survival in aquatic environment, possible pathways of contamination of aquaculture systems, serovars found in seafood and salmonellosis associated with fish and fishery products. The experts agreed that although *Salmonella* is a major foodborne pathogen, products of aquaculture are rarely involved in outbreaks of salmonellosis and the serovars which have been reported in products of aquaculture are rarely reported in cases of human salmonellosis in fish importing countries. The experts recognized that there are a variety of pathways reported as to how *Salmonella* can enter the aquaculture environment ranging from wild animals, domestic stock, poor sanitation and inappropriate disposal of human and animal wastes. Control of such pathways poses major challenges such as land runoff during rains, control of wild animals in the farm environment. There was agreement that very low level prevalence of *Salmonella* can be seen in products from aquaculture systems in developed countries but this has not led to any particular public health problem in these countries. The experts agreed that good hygienic practices during aquaculture production and biosecurity measures can minimize but not eliminate *Salmonella* in products of aquaculture. Biosecurity and control measures that would be useful in minimizing the risk of *Salmonella* contamination of aquaculture products were identified. The experts identified data gaps and made a series of recommendations to the national governments, national competent authorities, aquaculture industry and FAO.

CONTENTS

1. OVERVIEW OF THE EXPERT WORKSHOP

1.1 Introduction

International trade in aquaculture products has considerably increased in recent decades and the expansion of aquaculture production, particularly from Asia, has the potential to meet most of the growing global demand for fish and fishery products. The need to increase food production to feed an ever growing world population is more urgent now than ever as FAO has predicted that global food production will need to increase by 70 percent over current levels by 2050 (FAO, 2009a).

In addition to contributing to global food production, aquaculture is a major economic activity and an important source of foreign exchange for several developing countries. Currently aquaculture supplies about 50 percent of the global demand for fish and fishery products with about 90 percent of the aquaculture products coming from the Asian region (FAO, 2009b).

Ensuring food safety to protect public health and promote economic development remains a significant challenge for many countries worldwide. The application of risk-based approaches to food safety management is fundamental to international trade and underpins international food trade agreements. Harmonization of global trading standards relies on the application internationally agreed risk-based scientific principles which form the basis of the recommendations and standards of the Codex Alimentarius Commission. In the international market of fish and fishery products a major challenge faced by exporters is that different standards and regimes are applied by importing countries on producing countries to ensure that products meet the requirements of the target market.

Significant progress has been made in recent decades in improving the standards of post-harvest handling and processing of fish and fishery products with the introduction of food safety management systems based on the principles of the Hazards Analysis and Critical Control Point (HACCP) system. While food safety and hygiene standards have improved in the processing and manufacturing sectors, more still needs to be done to improve such food safety standards in the aquaculture production sector.

In recent years both FAO and WHO have advocated a risk based approach in food safety management and a current priority for both organizations is to promote risk based approaches to food safety management options in aquaculture production. Fish farmers are one of the first links in the food production chain and standards of final products depend on the application of good aquaculture practices being applied on the farm. A major challenge faced by many countries exporting aquaculture products is reducing the incidence of rejection of products because of microbiological contamination.

The presence of *Salmonella* remains a major cause of detention and rejection of shipments of raw aquaculture products in export markets. Though outbreaks of salmonellosis linked to products of aquaculture are rare, there is a concern in some importing countries that products coming from developing countries could be a source of *Salmonella* and this is exacerbated by detection of *Salmonella* in some raw fish and fishery products in import testing laboratories. Significant numbers of detections of this pathogen in fish and fishery products indicate that current strategies for *Salmonella* control in the aquaculture production and processing sectors are not adequate.

Our understanding of *Salmonella* ecology and reservoirs in various wild animals has been improving with number of studies looking at survival of *Salmonella* in rivers, soil and sources of contamination for aquatic environment. Application of microbial typing techniques such as serotyping and molecular typing has enabled better tracing of the source of pathogens. Outbreaks of salmonellosis associated with fresh produce have led to studies on presence and survival of *Salmonella* in irrigation water, which would be important consideration for aquaculture.

The objectives of this Expert Workshop were to:

i. review the current scientific knowledge regarding *Salmonella* ecology in aquatic environments, animal reservoirs and gain a better understanding of the pathways of contamination for aquaculture systems;
ii. consider epidemiological data on salmonellosis associated with fish and fishery products and facilitate application of risk based principles in the management of *Salmonella* problem in aquaculture;
iii. develop recommendations based on biosecurity and other measures for minimizing the contamination of products of aquaculture with *Salmonella*.

It is focused on the need for adopting novel biosecurity measures to minimize contamination and for using risk-based approach to develop management strategies for *Salmonella* in primary production systems to improve safety of fish for human consumption.

1.2 Summary of discussions and conclusions

The broad conclusions from the discussions of the workshop were:

(a) Public health risks due to *Salmonella* in products of aquaculture

- Although *Salmonella* is a major foodborne pathogen, products of aquaculture are rarely involved in outbreaks of salmonellosis.
- Serovars which have been reported in raw products of aquaculture are rarely reported in cases of human salmonellosis in fish importing countries.
- Very low level prevalence of *Salmonella* can be seen in raw products from aquaculture systems in developed countries but this has not led to any particular public health problem in these countries.
- This low level of prevalence may pose human health risk if aquaculture products are consumed raw, but even when low level of *Salmonella* is present, thorough cooking will eliminate the risk.
- While some marine fish caught offshore and handled hygienically and at low temperature according to the Codex Code of Practice for fish and fishery products (CAC/RCP/52-2003) may be suitable for raw consumption, it would be advisable to consume products of aquaculture only after cooking.

(b) Pathways of contamination of aquaculture environments

- There is a variety of pathways reported as to how *Salmonella* can enter the aquaculture environment ranging from wild animals, domestic stock, poor sanitation and inappropriate disposal of human and animal wastes. Control of such pathways pose major challenges such as land runoff during rains and, control of wild animals in the farm environment.
- *Salmonella* has been reported to survive for long periods of time in the aquatic tropical environment where aquaculture production takes place.
- There is currently disagreement in scientific literature as to whether or not *Salmonella* species are a part of the normal flora in the tropical aquatic environment. While long-term survival of some *Salmonella* serovars has been reported in these environments, it is unclear if these species are growing and multiplying and found an ecological niche. In some studies, the serotypes isolated from the environment are not those associated with humans and animals.

(c) Risk management

- Good hygienic practices during aquaculture production and biosecurity measures can minimize but not eliminate *Salmonella* in products of aquaculture.
- Current microbiological standards for fish and fishery products are based on fish from marine environment where pathogens like *Salmonella* are not expected to be present. Aquaculture is carried out in brackish water and inland freshwater environments, where opportunities for contamination with enteric organisms are higher.
- Good hygienic practices will minimize the opportunity for cross contamination in processing environments.
- Currently there is insufficient data to carry out quantitative risk assessment for *Salmonella* in aquaculture.

1.3 Data gaps and limitations

i. Product category-specific epidemiological data were only available for foodborne outbreaks in a proportion of developed countries. Further development of epidemiological surveillance systems should include the collection of such data on a wider basis.

ii. Comparison of the serovars isolated from seafood and the envrionment, and from seafood-associated human infections, infers that serovars may differ markedly in their potential to infect humans. The acquisition of further data on this aspect would inform potential future quantitative risk assessments and also inform hygiene controls and *Salmonella* standards for foods.

iii. The current dose response curves are determined using a variety of other foods. Studies on dose response using seafoods as the matrix using *Salmonella* isolates from seafoods and dose response information from outbreak data would be useful for quantitative risk assessments.

iv. There is very little quantitative data on *Salmonella* in various foods including seafoods. Exposure assessments would benefit from quantitative data at primary production and at the point of consumption.

1.4 Recommendations

To FAO

There are currently no internationally recognized guidelines for the safe production of aquaculture products. It is recommended that FAO work with key stakeholders to further develop these control measures into global guidelines for the safe production of aquaculture products.

FAO should work to harmonize the existing aquaculture certification systems.

To national governments

National governments should ensure that their food laws and regulations address the safe production of aquaculture.

National governments as a priority should implement an official food control programme that ensures the safe production of aquaculture products.

National governments should also ensure that adequate financial resources are available to implement the official food control programme for the safe production of aquaculture products.

To national competent authorities

National competent authorities should have a specific programme in place to minimize *Salmonella* contamination of aquaculture products.

National competent authorities should ensure that all staff working in the official food control programme working to minimize *Salmonella* contamination of aquaculture products should be adequately trained to allow them to perform their duties in a competent and consistent manner.

To the aquaculture industry

The aquaculture industry should assume responsibility for the production of safe aquaculture products and should implement a food safety management programme on the farm.

2. SALMONELLA: CHARACTERISTICS AND PUBLIC HEALTH OUTCOMES

2.1 General characteristics and association with human infections

Salmonella is a facultatively anaerobic, Gram-negative bacterium that can cause illness in humans and other animals. Most strains are motile by means of flagella. There are formally two species of *Salmonella*, *Salmonella enterica* and *Salmonella bongori*. Both species are divided into serovars. There are six subspecies of *S. enterica*: *S. enterica* subsp. *enterica*, *S. enterica* subsp. *salamae*, *S. enterica* subsp. *arizonae*, *S. enterica* subsp. *diarizonae*, *S. enterica* subsp. *houtenae* and *S. enterica* subsp. *indica*. The strains that cause human illness are primarily included in the subspecies *S. enteric*a subsp. *enterica*.

Salmonella are excreted in the faeces of animals (including birds) and humans that are infected with, or asymptomatically excreting, the organism. A biovar (previously known as S. Java) of *S.* Paratyphi B, strains of *S. enterica* other than subspecies *enterica*, and *S. bongori* are associated with cold-blooded animals such as amphibians, reptiles, although at least one serovar of *S. enterica* subsp. *diarizonae* has been found in sheep. Human infections have arisen from contact with both turtles and frogs kept in aquaria – the latter has included infection with *S.* Typhimurium (CDC, 2010). *Salmonella* has also been reported as being present in the guts of river fish (Gaertner *et al.*, 2008). Those workers suggested that cold blooded animals such as turtles provide habitats that allow the persistence of *Salmonella* in the environment but that they do not play a significant role in the dissemination of *Salmonellae* in the natural environment. It has also been put forward that *Salmonella* may be part of the normal flora in aquatic environments, at least in tropical regions (Reilly and Twiddy, 1992).

Transmission of *Salmonella* to humans is predominantly via water or food contaminated with faecal material, or cross-contaminated from other products containing the organism, or contaminated by infected food-handlers. Insects can also carry *Salmonella* in their gut and can contaminate food. In some types of birds, vertical transmission occurs from the female to the egg and humans can become infected by eating the latter. In developing tropical countries, the waterborne route predominates while in countries with better general public health the foodborne route is more important.

Salmonella strains contain a number of somatic ("O") and flagellar ("H") antigens. Many strains may, at any one time, express one of two different flagellar compositions, which give rise to two "H" phases. Assignment to serovar necessitates identification of the "O" and "H" antigens, including both phases if this is relevant. Other antigens may also be present, for example: Vi in some strains of *S.* Typhi, *S.* Paratyphi C and *S.* Dublin. Approximately 2 500 serovars have been described. Only a relatively small proportion predominates in reported human infections. Ninety nine percent of the human infections are due to *Salmonella enterica* which has about 1 500 serotypes. Based on an analysis of globally reported food borne outbreaks, the non-typhoid *Salmonella* serotypes most often encountered in human infections are Enteritidis followed by Typhimurium (Greig and Ravel, 2009). In a broad review of serovars reported from human infections, the following were the ten most commonly found: *S.* Enteritidis, *S.* Typhimurium, *S.* Hadar, S. Infantis, S. Newport, S. Typhi, *S.* Agona, *S.* Virchow, *S.* Heidelberg and *S.* Derby (Herikstad *et al.*, 2002). However, the predominant serovars found in human infections vary both geographically and with time. For example, *Salmonella* Weltrevreden was the second most common serovar in Asia during 2000-2001, but this serovar dropped to fourth place in 2002 surpassed by *S.* Rissen and *S.* Typhimurium (Galanis *et al.*, 2006).

S. Typhi is a common cause of human infection in Africa and South-East Asia, but less common elsewhere. Some of the common serovars, such as S. Enteritidis, S. Typhimurium and S. Virchow, as well as S. Typhi and S. Paratyphi, can be further subdivided using phage-typing.

The clinical outcomes of *Salmonella* can be considered as two separate groups:

Salmonella Typhi/Paratyphi – Strains of these cause enteric fever, a serious systemic illness. Incubation period ranges from 7 to 28 days. Symptoms include malaise, headache, fever, cough, nausea, vomiting, constipation, abdominal pain, chills, rose spots, bloody stools.

Non-typhoid *Salmonella* – Strains of these may cause gastroenteritis in humans. Incubation period ranges from 8 to 72 hours. Symptoms include abdominal pain, diarrhoea, chills, fever, nausea, vomiting and malaise. Systemic infection such as septicaemia may occur especially in susceptible patients such as the very young, very old and immunocompromised. The available data measuring illness as the endpoint suggests that no response is observed until a dose of 10^6 is reached (Coleman and Marks, 1998). Severe dehydration due to diarrhoea can on occasion require medical intervention through the administration of intravenous fluids and antibiotic treatment. However, occasionally some serovars of this pathogen may cause sepsis after entering the blood stream from the intestine and require intense medical intervention. Death is rare if patient is promptly hydrated and provided antibiotic treatment.

2.2 Pathogenicity and host factors

2.2.1 Host factors

Salmonella species are a leading cause of food borne illnesses worldwide and their incidence is dependent upon a variety of factors including host susceptibility. In general, the host factors that can affect outcome of exposure to *Salmonella* or any food borne pathogen by ingestion include age, nutritional status, socio-economic and environmental factors, immune status and underlying diseases. This susceptibility can often be associated with socio-economic status and demography.

Age and the general health of the exposed population are factors that should be considered when assessing the susceptibility of the host to infection. In addition to age, the immunological condition of the host apparently plays a significant role in disease. Children who have immature immune systems and people who are immuno-compromised show increased susceptibility to salmonellosis (FAO/WHO, 2002). It has been noted that children who have more neutral stomach pH are more susceptible due to the acid susceptibility of these pathogens. While stomach pH can affect host susceptibility, the matrix in which the pathogen is consumed may promote/protect the agent from low pH in the stomach. Following resolution of the acute phase, excretion of *Salmonella* ceases within several weeks, although a carrier state may evolve.

2.2.2 Pathogenicity factors

Pathogenicity of non-typhoid *Salmonella* strains is influenced by the presence of several pathogenicity islands in the genome – referred to as *Salmonella* pathogenicity islands (SPI). At present 12 different SPI have been described. Additional pathogenicity factors may be located on a plasmid. The islands and plasmid contain genes influencing attachment, invasiveness, production of toxins, and the survival and growth in the host.

A variety of fimbrial adhesins are involved in initiation of contact to host cells (Hensel, 2004). While the roles in pathogenesis of some SPI are well defined, the function in virulence of many genes within SPI are not understood (Hensel, 2004). The O side chains of the lipopolysaccharide molecules have also been shown to affect invasiveness and enterotoxin production (Murray, 1986). Other factors that affect the ability of the organism to cause disease include the presence of cytotoxins and diarrhoeagenic enterotoxins. The enterotoxin is released into the lumen of the intestine and results in the loss of intestinal fluids (D'Aoust, 1991). Antimicrobial resistant strains are somewhat more virulent than susceptible strains, in that, they cause more prolonged or more severe illness than do antimicrobial susceptible strains (Travers and Barza, 2002).

2.2.3 Effect of the food matrix on infectivity

Gastric acidity (pH 2.0) is considered an important defence against food borne pathogens. Though normally *Salmonella* grow at neutral pH, they have complex survival strategies that might facilitate their ability to tolerate pH fluctuations during pathogenesis. Most aquaculture products have neutral pH and *Salmonella* may be protected in this food matrix. The outcome may be affected by the amount of food ingested, the fat content and buffering capacity of the food, and the nature of contamination. In one outbreak linked to the consumption of scallop with egg yolk, 6.30 log cells resulted in a 56 percent attack rate (FAO/WHO, 2002). In fatty foods (e.g. chocolates, cheddar cheese) low infective dose may be observed and some aquacultured fish (e.g. salmon, catfish) may have high fat content, but there is no available data on outbreaks associated with these fish. Increased attack rates have been associated with ingestion of *Salmonella* between meals and it has been postulated that pyloric barrier may fail at this time and chocolates and ice creams may be consumed between meals (Mossel and Oei, 1975). Distribution of bacteria in food may also affect the outcome and due to the nature of bacteria to grow in colonies, agglomeration of cells may occur in foods and cells in inner layers of this might be protected (FAO/WHO, 2002).

2.3 Dose-response relationships

Nine studies have been published of experimentally induced salmonellosis, conducted between 1936 and 1970 using a variety of serotypes and strains (Table 1). However, some of these studies were deemed to be unsuitable to derive conclusions about the pathogenicity of *Salmonella* in general in humans. Severe illness resulting from salmonellosis can be exacerbated by antibiotic resistant strains of *Salmonella* and may be further complicated by the effects of other underlying illnesses.

There are number of human feeding trials performed using six different serotypes (Table 1). There were usually no illnesses at doses less than 10^6. However, outbreak investigations show that lower number of cells can cause infection depending upon the food matrix. There is no data with sea food matrix alone but in an outbreak of *S.* Enteritidis associated with scallop and egg yolk, a 56 percent attack rate was observed at a dose of 6.3 log Colony Forming Units (CFU). More information on the outbreaks, attack rate and doses involved is available in Table 3.14 of the FAO/WHO risk assessment of *Salmonella* in eggs and broiler chicken (FAO/WHO, 2002). Future risk assessments would benefit from data on levels of *Salmonella* in seafoods involved in any outbreaks.

Table 1. Human feeding trials using *Salmonella* (FAO/WHO, 2002)

No.	Serotype(s)	Strain(s)	References
1	S. Typhimurium		Hormaeche, Peluffo and Aleppo, 1936
2	S. Anatum		Varela and Olarte, 1942
3	S. Meleagridis S. Anatum	I, II & III I, II & III	McCullough and Eisele, 1951a McCullough and Eisele, 1951a
4	S. Newport S. Derby S. Bareilly		McCullough and Eisele, 1951c McCullough and Eisele, 1951c McCullough and Eisele, 1951c
5	S. Pullorum	I, II, III & IV	McCullough and Eisele, 1951d
6	S. Typhi		Sprinz *et al.*, 1966
7	S. Sofia S. Bovismorbificans		Mackenzie and Livingstone, 1968
8	S. Typhi	Quailes, Zermatt, Ty2V, 0-901	Hornick *et al.*, 1970
9	S. Typhi	Quailes	Woodward, 1980

3. SALMONELLA IN THE AQUATIC ENVIRONMENT

3.1 Occurrence in the aquatic environment

The presence of *Salmonella* in the external environment and food is considered a critical step to ensure the passage of the bacteria to new hosts. Existing studies on the presence of *Salmonella* in aquatic systems and seafood products have identified two different patterns according to the climate characteristics of the area. The results of a number of studies are summarized in Table 2. In different temperate and arid regions of Spain (Martinez-Urtaza *et al.*, 2004), Morocco (Setti *et al.*, 2009) and Mexico (Simental and Martinez-Urtaza, 2008) characterized by of presence of cold coastal seawaters, *Salmonella* have been detected in less than 10 percent of seawater and shellfish samples investigated. Similar low levels of *Salmonella* were obtained in marine samples from regions with similar oceanographic conditions to these areas and with temperate seawater temperatures, such as the United States and the United Kingdom, which have incidences of 7.4 percent, and 8 percent, respectively (Brands *et al.*, 2005; Martinez-Urtaza *et al.*, 2004; Wilson and Moore, 1996). These results contrast with the high prevalence in tropical areas where *Salmonella* incidence in seafood can reach up to 20 percent of the samples, as it has been reported for areas of Asia and Africa (Hatha and Lakshmanaperumalsamy, 1997; Heinitz *et al.*, 2000). In Vietnam, an incidence of 18 percent of positive samples for *Salmonella* has been reported for shellfish product (Van *et al.*, 2007), whereas in India, presence of *Salmonella* was found in 24.3 percent of different seafood products investigated (Rakesh Kumar *et al.*, 2008a). A recent study of presence of *Salmonella* in rivers and coastal waters carried out in tropical areas of Mexico has shown an occurrence of this organism over 80 percent (Jimenez, Chaidez and Martinez-Urtaza, personal communication). A total of 75 percent prevalence has also been reported in Oconee river basin, Georgia, USA (Meinersmann *et al.*, 2008).

The dynamics of contamination of *Salmonella* in natural environments has been associated with specific seasonal patterns or climate characteristics. In temperate and tropical regions, presence of *Salmonella* in the environment has been detected linked to the periods of rains, and more specifically, after the days of the first heavy rains. Therefore, associations between storm-generated flows, torrential rains, and the monsoon season have been reported in studies in temperate and tropical regions of the world with frequent rainy periods (Baudart *et al.*, 2000; Brands *et al.*, 2005; Hatha and Lakshmanaperumalsamy, 1997; Martinez-Urtaza *et al.*, 2003; O'Shea and Field, 1991; Venkateswaran *et al.*, 1989), signalling the washing effect of torrential rains as one of the principal environmental drivers of *Salmonella* contamination in coastal areas (Martinez-Urtaza *et al.*, 2003). The arrival of *Salmonella* into marine environments is predominantly governed by the presence of persistent rains enough to transport the contamination from the original source points to the sea via aquifers, streams and rivers (Simental and Martinez-Urtaza, 2008). *Salmonella* has been found in several marine mammals like porpoises, sea lions, whales, dolphins, seals (Higgins, 2000). The permanence of *Salmonella* contamination in aquatic and marine systems appears to have been modulated by a combination of oceanographic characteristics and atmospheric conditions related primarily to the effects of sunlight. The presence of intense sunlight has been identified as a critical factor for the drastic reduction of *Salmonella* spp. in coastal areas of Spain, Morocco and Mexico (Martinez-Urtaza *et al.*, 2004; Setti *et al.*, 2009; Simental and Martinez-Urtaza, 2008). Water temperature has been proposed as playing an important role in the long-term survival of *Salmonella* in the environment. The presence of cold waters may reduce the survival of *Salmonella* spp. in the marine environment, while warm waters together with high levels of organic matter, typical conditions prevailing in tropical areas, may contribute to a more appropriate habitat for an increased survival of bacteria, as reflected in the disparate incidence of *Salmonella* described in diverse studies in temperate and tropical regions. *Salmonella* has been detected prevailing all year round in tropical areas in a recent study performed in Sinaloa, Mexico (Jimenez, Chaidez and Martinez-Urtaza, personal communication).

Table 2. *Salmonella* detection in the aquatic environment

Country (No of samples)	Type of sample	Positive sample (%)	Serotypes	Resistance (%)	Reference
Spain (5384)	Molluscs	3	Serotypes (N=20): Senftenberg 42.5% Typhimurium 15% Agona 9.4%	9	Martinez-Urtaza *et al.*, 2004
	Seawater	2.5			
Morocco (801)	Mussels	10	Serotypes (N=3): Blockley 43.8% Kentucky 29.8% Senftenberg 26.3%	49.1	Setti *et al.*, 2009
	Sediments	6.8			
	Seawater	4.1			
Mexico, Ensenada (1331)	Wastewater	16.2	Serotypes (N=20): Typhimurium 23.4% Vejle 6.2% Suberu 4.7%	–	Simental *et al.*, 2008
	Stream water	10.6			
	Molluscs	7.4			
	Seawater	2.3			
Mexico, Culiacan (138)	Water	80.4	Serotypes (N=29): Oranienburg 24,3% Saintpaul 9.0% Minnesota 6.3%	50.4	Jimenez, Chaidez and Martinez-Urtaza, personal communication
Asian countries (1234)	Shrimps	1.6	Weltevreden Paratyphi B Abaetetuba	–	Koonse *et al.*, 2005
	Holding pond water	2.5			
	Pond sediments	1.0			
	Pond grow-out water	3.5			
	Source water	5.0			
	Source sediment	24			
Viet Nam (50)	Shellfish	18.0	-	11.1	Van *et al.*, 2007
India, Cochin (443)	Fish	30.5	Serotypes (N=30): Weltevreden 8.2% Rissen 7.8% Typhimurium 6.7%	82%	Kumar *et al.*, 2008
	Shrimps	29			
	Clams	34.1			

The vast majority of studies looking at the presence of *Salmonella* in aquatic and marine environments have evidenced two main observations: only a small but constant number of serovars have been found in these environments and, in most cases, these do not coincide with the main zoonotic serovars identified in the surrounding areas (Catalao Dionisio *et al.*, 2000; Heinitz *et al.*, 2000; Martinez-Urtaza *et al.*, 2004; Polo *et al.*, 1999; Venkateswaran *et al.*, 1989; Wilson and Moore, 1996). In spite of the variability in sampling size (n= 37 to 251), in most of these studies the maximum number of serotypes identified has been around 20 (Catalao Dionisio *et al.*, 2000; Martinez-Urtaza *et al.*, 2004; Venkateswaran *et al.*, 1989; Wilson and Moore, 1996). Serovar Typhimurium has been shown to be the most common clinically significant serovar isolated from environmental samples in many parts of the world (Baudart *et al.*, 2000; Catalao Dionisio *et al.*, 2000; Martinez-Urtaza *et al.*, 2004; Polo *et al.*, 1999; Willson and Moore, 1996; Simental and Martinez-Urtaza, 2008), which attests to its capacity of adaptation and survival in external environments (Baudart *et al.*, 2000). *Salmonella* Senftenberg has been recognized one of the major serotypes identified in marine environments and raw seafood worldwide. It has been one of the predominant serovars detected in the coastal waters of Portugal (Catalao Dionisio *et al.*, 2000), in crustaceans from India (Hatha and Lakshmanaperumalsamy, 1997), in raw seafood imported into the United States especially from tropical countries (Heinitz *et al.*, 2000),