

A Review of *Salmonella* sp. in Tilapia fish (*Oreochromis niloticus*) : Public Health Importance

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ABSTRACT

Tilapia fish or *Oreochromis niloticus* is a food security commodity from the fisheries sector and is traded internationally, but in Asian countries tilapia aquaculture is mostly maintained using traditional systems, besides that homemade feed is used to reduce production costs. Cultivation of tilapia with traditional systems and artificial feed that is integrated with other livestock has the potential to cause the danger of transmitting zoonotic pathogens from other livestock manure and feed residue. *Salmonella* sp. which is one of the zoonotic pathogens that can be transmitted from tilapia. The use of antibiotics in livestock and cultivation causes *Salmonella* sp. able to withstand some antibiotics. Salmonellosis in humans has become an important public health problem, incurring significant economic and medical costs worldwide. The incidence of salmonellosis due to fish consumption has become a concern of public health agencies in several countries, because increased consumption of fishery products, especially raw products, increases the risk. Pathogen exposure, especially in vulnerable groups, such as the elderly, has increased significantly. pregnant women and babies. The importance of this pathogen in fish can be assessed and evaluated, as records show that most *Salmonella* infections in humans are related to fish consumption.

Keywords: Tilapia fish, *Salmonella* sp, Zoonotic pathogen, Public health

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INTRODUCTION

Integrated tilapia farming is a traditional practice by small-scale farmers in China as well as in other Asian countries. Fish are usually raised in ponds with livestock units, such as pigs, located on pond embankments, which allow drainage of livestock manure and excess feed into the pond as plankton and other fish feed [1, 2]. Although integrated agricultural systems are sustainable in many ways, they represent potential food safety hazards, for example transmission of zoonotic pathogens from faeces and accumulation of antimicrobials and others. residues originating from pig manure and feed [3, 4]. Tilapia is one of the easiest to cultivate and trade fishery food products internationally in the world, with an estimated 1.45 million tonnes produced in China in 2013 [5, 6].

In the Asia-Pacific region, farmed fish are fed commercial or homemade feed [7]. Homemade feed is used to reduce costs and usually consists of chicken offal and by-products produced during poultry processing, kitchen waste, and other by-products from the food industry [7, 8]. Homemade diets can be a potential source of foodborne pathogens, particularly *Salmonella* bacteria [9, 10], which can then be transmitted to farmed fish [11] and to humans. Salmonellosis in humans has become an important public health problem, creating significant economic and medical costs worldwide. In the United States, nontyphoidal *Salmonella* caused approximately 1 million foodborne illnesses, 13,000 hospitalizations, and 242 deaths in 2011 [12]. In Taiwan, *Salmonella* is one of the most frequently isolated foodborne pathogenic bacteria [13]. *Salmonella* is usually transmitted to humans through consumption of

contaminated food products. According to Bean et al. [14], approximately 7% of cases of human salmonellosis result from ingestion of contaminated food from fishery products, particularly ready-to-eat (RTE) products. In Hong Kong, detected *Salmonella* in smoked fish, salmon sushi, and salted fish products [15, 16, 17], which illustrates that *Salmonella* was continuously contaminated by these RTE products. In addition, human salmonellosis caused by contaminated fish products has been reported in European countries [18, 19], where processing is considered to be one of the important sources of contamination [20, 21]. Martinez-Urtaza and Liebana [21] and Morris et al. [22], who evaluated the incidence of *Salmonella* in seafood processing plants, surveillance of foodborne pathogens during processing is essential to a successful contamination control program. When monitoring *Salmonella* contamination, serotyping and molecular subtypes are often applied to trace the source of bacterial contamination [23, 24].

The incidence of salmonellosis due to fish consumption has become a concern of public health agencies in several countries, because the increased consumption of fish farming, especially raw products, which increases the risk of exposure to pathogens, especially in vulnerable groups, such as the elderly, increases significantly. pregnant women and infants [25, 26]. The importance of this pathogen in fish can be assessed and evaluated, as records show that fish is the source of transmission responsible for 7% of total foodborne outbreaks in Europe in 2016, with the majority of *Salmonella* infections in humans linked to

fish consumption caused by *Salmonella* Typhimurium and Enteritidis serovars [27, 28, 29, 30].

Overview of *Salmonella* sp.

Salmonella spp. are Gram-negative rod-shaped bacteria that cause salmonellosis. In humans, these pathogenic bacteria cause enteric fever and acute gastroenteritis [31]. Symptoms include mild to severe gastroenteritis, with an incubation period of 6–72 hours [31]. Salmonellosis outbreaks due to fish consumption have been reported in several countries. For example, salmonellosis caused by eel consumption, which is linked to fish farming in Italy and has been reported in Germany [32]. Several reports have linked the presence of *Salmonella* spp. in various fishes and shellfishes [33, 34]. Various hazards associated with natural fish farming come from the environment or from human or animal activities. Fish can be a vehicle for transmission of *Salmonella* which can be pathogenic for humans and has a high potential to transmit its antibiotic resistance gene to other pathogens via plasmids [35]. The potential for antibacterial agents to cause the development of resistance in fish pathogens is of worldwide concern [36].

Salmonella grows optimally at 37 °C, but these bacteria are able to grow in a temperature range of 5–47 °C [37]. However, dietary *Salmonella* exhibits varying survival behavior depending on the type of food matrix and storage conditions [37]. Studies show that *Salmonella* can survive for more than 16 weeks in frozen processed chicken products and for more than 9 months in frozen beef without much change in quantity [37]. Studies on the viability of *Salmonella* in chilled and frozen fish and fishery products are very limited. One study reported that *Salmonella* Enteritidis survived but did not grow at 3 °C, whereas bacteria thrived at 10 °C in vacuum packed fish and poultry [38]. The amount of *Salmonella* in raw tuna was reduced by 1 to 2 logs after 12 days of storage at 5–7 °C, whereas in frozen storage *Salmonella* became undetectable after 42 days of storage. In frozen shrimp, populations of *S. Weltevreden* and *S. Senftenberg* decreased but were not eliminated during 12 weeks of storage [39, 40].

Zoonoses in humans

Infection can occur if fishery food contaminated with salmonella is consumed raw or undercooked. Salmonellosis transmitted through food from fisheries is a major cause of human morbidity worldwide [40, 41]. *Salmonella* spp. cause various diseases such as enteric fever and gastroenteritis. The majority of the estimated 1.3 billion annual cases of salmonellosis result from consumption of contaminated food products especially pork, poultry, eggs and vegetables [41]. In comparison, fishery foods are less frequently associated with *Salmonella* infections although reports from Thailand, Malaysia and other Asian countries indicate that certain serovars, eg *S. Weltevreden*, may be primarily associated with aquatic environments and seafood [42]. Prevalence of *Salmonella* spp. may also be higher in seafood from Asia compared to seafood from other geographic regions [43, 44]. However, other studies argue that *Salmonella* spp. can last longer in tropical aquatic environments and should therefore be seen as part of the normal micro-flora of aquaculture products of the area [45]. *Salmonella* can be disseminated as a result of water currents, underground springs and rainwater runoff carrying contaminated material [46–49]. Like *E. coli*, *Salmonella* is continuously released into the environment from infected humans, livestock, domestic animals, and wildlife [50]. Pathogenic and potentially pathogenic bacteria associated with fish

and shellfish include *Mycobacteria*, *Streptococcus iniae*, *Vibrio vulnificus*, *Vibrio* spp, *Aeromonads*, *Salmonella* spp, *Shigella* and others [51, 52, 53]. Human infection by these fish pathogens is usually through contact with infected fish during handling, water or other elements of the fish's living environment [54]. The initial microflora on the surface of the fish is directly related to the aquatic environment whereas the flora in the digestive tract is related to the type of food and condition of the fish [55]. *Salmonella* sp. is a causative agent of pathological diseases in humans, cattle, poultry and other livestock and *Salmonella* infection is a major cause of concern for humans, animals and the food industry. Characterization of more than 2,500 *Salmonella* serovars, the pathogen infects nearly all vertebrates but the severity of infection varies from one serovar to another depending on the specificity of the host [56]. Some *Salmonella* serovars are limited to one or several hosts while others have a wide spectrum of hosts. An understanding of the mechanisms involving host preference by one serovar over another is essential. This is used to increase our knowledge about host adaptability and will be instrumental in designing better prevention models. Methods involving identification of genetic markers for host specificity will prove to play a role in determining virulence factors for other bacterial pathogens that cause systemic infection [54, 56].

The low standard of health in the coastal areas increases the eutrophication process, thus creating a broad conducive environment for the survival of microbes that eventually infect fish. Analysis of fish tissue slurry showed that the fish harvested from the landing beaches along the coast that had been contaminated by Enterobacteriaceae were; *Salmonella*, *Shigella* and *E. coli* [57, 58, 59]. In the Asia-Pacific region, farmed fish are fed commercial or homemade feed [60]. Homemade feed is used to reduce costs and usually consists of chicken offal and byproducts produced during poultry processing, kitchen waste, and other by-products from the food industry [60, 61]. Homemade diets can be a potential source of foodborne pathogens, particularly *Salmonella* bacteria [9, 62], which can then be transmitted to farmed fish [63, 64] and, in turn, to humans. Detailed information on salmonellosis due to consumption of tilapia in Malaysia is lacking, as most cases of food poisoning are not reported to the authorities. However, the Malaysian National Public Health Laboratory reports that the five most common nontyphoidal *Salmonella* serovars are *Salmonella* Enteritidis, *Salmonella* Weltevreden, *Salmonella* Corvallis, *Salmonella* Typhimurium, and *Salmonella* Tshiongwe [65]. In Malaysia, freshwater fish are kept using aquaculture ponds, ex-mining ponds, freshwater cages, cement tanks, canvas tanks, and freshwater cage culture systems. Most of the freshwater cultivated fish are maintained using the aquaculture system (59.5%) and ex-mining ponds (25%). Catfish (58.1%) and tilapia (41.3%) were reared in ground ponds and ex-mining [65].

Transmission of bacteria from tilapia (*Oreochromis* sp.)

Fish is considered to be one of the most nutritious and highly desirable food ingredients due to its high nutritional value and is rich in protein, vitamins and unsaturated fatty acids. Fish contains n-3 polyunsaturated fatty acids which is a very important aspect for health-conscious people especially in affluent countries where the death rate from cardiovascular disease is high. Fish contains more easily digestible protein than protein found from other sources. Fish are of great concern for export

earnings because of their higher nutritional value, low cholesterol levels and presence of essential amino acids [64]. Bangladesh is the 5th in world aquaculture production, which accounted for half of the country's total fish production in 2015-2016 [65, 66]. This sector contributed 3.65% to GDP and 1.97% to foreign exchange earnings through exports [66]. It is the second largest export industry in Bangladesh and accounts for 2.5 percent of global shrimp production [66].

The main pathogens affecting cultivation include bacteria, fungi, viruses, and parasites [67-69]. Bacterial diseases have become a major concern for cultivation, especially with warm water temperatures [70]. Different bacterial species are reported to be pathogens against aquatic tilapia, including *Aeromonas hydrophila*, *Edwardsiella tarda*, *Flavobacterium columnare*, *Francisella* spp., *Yersinia ruckeri*, *Staphylococcus epidermidis*, *Vibrio vulnificus*, and *Streptococcus agalactiae* [71-79].

Fish was associated with 24% of foodborne disease outbreaks and 6% of all food poisoning, or foodborne illness [80]. Levels of pathogenic bacteria in tilapia are related to the environment and its handling before entering food markets and restaurants. The bacteria associated with tilapia fish can be transmitted by direct contact and cause foodborne illness. For example, handling of tilapia has been linked to an outbreak of *Vibrio vulnificus* in a Seattle supermarket [81]. Other foodborne pathogenic bacteria including *Salmonella enterica*, enteropathogenic *Escherichia coli*, *Listeria monocytogenes*, *Yersinia enterocolitica*, and *Klebsiella pneumoniae* were identified from fresh tilapia in the Kenyan freshwater fish chain [82]. Shigatoxigenic and enteropathogenic *Escherichia coli* was isolated from farmed tilapia (*Oreochromis niloticus*) in the northeast region of the state of Sao Paulo [83]. It was reported that the microbial quality of tilapia showed that all the tissues were contaminated by salmonella and fecal coli. *Salmonella* can spread as a result of water currents, underground springs and rain runoff carrying contaminated material [84, 85]. Human infection by these fish pathogens is usually through contact with infected fish while handing them over, water or other elements of the fish's living environment [86]. These pathogenic organisms have been isolated from freshwater fish such as *Tilapia nilotica* Linn [87]. Tilapia is an important aquaculture production for food supply. Because on a global scale, fish and fish products are the most important sources of protein and it is estimated that more than 30% of the fish consumed by humans comes from aquaculture [88, 89].

Safe consumption of fish and fishery products requires adequate sanitary conditions from harvest to consumption [90, 91]. Consumption of fish and shellfish can also cause illness due to infection or poisoning. Most of the foodborne diseases are caused by *Salmonella* spp., *Staphylococcus* spp. and *Escherichia coli*, usually associated with the consumption of fish infected with these bacterial species especially *Salmonella* and *E. coli* [92, 93]. *Salmonella* usually enters the human intestine with food, *Salmonella* spp. must overcome resistance to colonization mediated by the gut microbiota and the innate immune system. *Salmonella* by inducing inflammation and innate immune defense mechanisms. Many models have been developed to study *Salmonella* spp. interactions with the microbiota have helped to identify the factors needed to overcome colonization resistance and to mediate disease [10, 54, 93]. *Salmonella* infection in humans mainly includes typhoid fever and this infection, known as enteric fever, continues

to be one of the most serious public health problems worldwide. The presence of higher levels of *Salmonella* in fish causes several symptoms in the human body such as diarrhea, nausea, vomiting and abdominal pain. Human salmonellosis caused by contaminated fish products has been reported in European countries [18, 19], where processing is considered to be one of the important sources of contamination [20, 21].

ANTIBIOTIC RESISTANCE

Self-limiting gastroenteritis is the main clinical picture developed by *Salmonella*, which in severe cases may require fluid and electrolyte replacement. The use of antibiotics is reserved for patients with serious illness or a high risk of invasive disease [94]. The antibiotic therapy scheme for typhoid fever includes third generation cephalosporin antibiotics, quinolones and macrolides. However, recently it has developed between typhoid *Salmonellas* and non-typhoid strains with a high degree of resistance to quinolones and cephalosporins [95]. The emergence of several drugs resistant to *Salmonella* (MDR) is currently of worldwide concern, and the occurrence of *Salmonella* MDR in food is a risk condition, indicating an increase in the severity of foodborne disease, leading to increased hospitalization rates and the likelihood of death [96]. In contrast, the epidemiology of antimicrobial resistance of *Salmonella* spp. It is complex and can be influenced by factors such as: consumption of antibiotics, human travel, transmission between patients in hospitals, import and trade in food of animal origin or not, trade in live animals within the country or between countries and exposure through animals or the environment human [97].

The use of antibiotics for therapeutics and growth promoters in feed animals has shown that the main factor for the emergence of resistant isolates [98], has been reported from livestock [99-103], poultry [104-108], pets [109-112], fish [113-116], and from animal products. [117-120]. Several studies have reported that pet food, meat, milk and fishery products contaminated by a multi-resistant *S. aureus* strain which has been one of the common causes of severe nosocomial infection for a long time [121]. In contrast, a large number of studies have reported increased incidence of resistance among *Salmonella* spp. isolated from poultry, beef and fishery products [122].

Antibiotics are used for medicinal purposes and as growth promoters in livestock and aquaculture leading to the development of resistance [123]. Acquired resistance to tetracyclines and chloramphenicol has been associated with extensive use of antibiotics in aquaculture in several Asian countries [124]. Sapkota et al. [125] reported that of the top 13 aquaculture producing countries, 92% used oxytetracycline and 69% used chloramphenicol.

It was reported that antibiotic resistance in Gram-negative bacteria from pond culture was higher in ponds undergoing antimicrobial therapy or with a recent history of treatment than in ponds without antimicrobial treatment recently [126, 127]. In a previous study, it was reported that all *S. enteritidis* strains isolated from tilapia sold in a wet market in Selangor, Malaysia were susceptible to rifampin. The emergence of *Salmonella* serovars with high MAR index indicates that these serovars originate from environments where antimicrobials are often used as therapy or as growth promoters in animal feed [128, 129]. Some drug-resistant *Salmonella* isolates have been suggested to be more virulent than non-multiple drug-resistant *Salmonella*

isolates [130]. *Salmonella* resistance to one or more antibiotics has been reported by many investigators [131]. Horizontal transfer of resistance genes in plasmids has been shown between bacteria in fish pond water [131] and in marine sediments [132]. Plasmids in *Salmonella* spp. has been reported to transfer antibiotic resistance and virulence properties [35].

CONCLUSION

The presence of higher levels of *Salmonella* sp in tilapia fish causes several symptoms in the human health such as diarrhea, nausea, vomiting and abdominal pain. *Salmonella* sp in tilapia obtained from the decay of animal waste and feed residue during the traditional cultivation process. *Salmonella* sp is often resistant to several antibiotics which allow it to be transferred to the surrounding environment, which is because in the process of tilapia pond cultivation it is mixed with disposal from livestock waste where there is a lot of antibiotic residue and while cultivation is still using antibiotics. This is what needs to be a public health concern about antibiotic resistance from isolates obtained in tilapia fish.

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