

Microbial Safety of Fish and Fishery Products

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1.0 Introduction

Microorganisms play a vital and distinct role in the safety of fish and fish products. Ensuring the safety of fish products necessitates the effective control of both pathogenic microorganisms and spoilage bacteria due to their potential effect on human health. Bacteria are found on fish skin, gills, digestive tracts, and organs including the kidney, liver, and spleen. Fish and fish products, particularly raw or undercooked ones, have caused outbreaks of bacterial infections, biotoxins, histamine, viruses, and parasites. Fish have been identified as reservoirs of various bacterial pathogens that have been linked to human diseases. These pathogens include *Mycobacterium spp.*, *Streptococcus iniae*, *Photobacterium damsela*, *Vibrio alginolyticus*, *Vibrio vulnificus*, *Vibrio parahaemolyticus*, *Vibrio cholerae*, *Erysipelothrix rhusiopathiae*, pathogenic *Escherichia coli*, *Aeromonas spp.*, *Salmonella spp.*, *Staphylococcus aureus*, *Listeria monocytogenes*, *Clostridium botulinum*, *Clostridium perfringens*, *Campylobacter jejuni*, *Delftia acidovorans*, *Edwardsiella tarda*, *Legionella pneumophila*, and *Plesiomonas shigelloides*. Furthermore, the existence of antibiotic-resistant genes (ARGs) within these microorganisms has raised concerns regarding the dissemination of antibiotic resistance (AMR) in both the environment and human populations. In addition to pathogens that impact human health, bacteria have been identified as the primary factor responsible for fish spoilage. Certain spoilage bacteria possess decarboxylase enzymes that can effectively convert free histidine into substantial quantities of histamine. The consumption of such food items can potentially result in scombroid poisoning, a significant concern in terms of food safety.

2.0 The infiltration of microorganisms:

In living conditions, when fish are first introduced to the environment, they are exposed to a wide range of microorganisms, but the fish's immune system keeps bacteria away from growing in the muscle of the fish. Once the fish dies, its immune system breaks down, letting bacteria grow out of control. During storage, they move between the muscle fibres and into the flesh. But, very few bacteria got into the meat when it was stored in ice. Since only a small number of organisms actually get into the flesh, and most microbial growth happens outside, spoilage is probably caused by bacterial enzymes getting into the meat and nutrients getting out. Still, the types of microbes usually found in fish and fish products fall into two groups: I.

microorganisms responsible for spoilage, II. microorganisms that cause disease, i.e., Pathogenic bacteria

3.0 Potential Spoilage bacteria of fish:

Fish and fish are often considered favourite foods due to their deliciousness, high protein content, unsaturated fatty acid content, and omega-3 fatty acid content. However, fish quickly spoils after being caught due to the biological and chemical components and microbial load. After twelve hours, the putrefaction process will start because of the metabolic activity of bacteria, the activity of endogenous enzymes (autolysis), and the oxidation of lipids caused by chemical reactions. After the catch, fish are particularly susceptible to spoilage, and it is essential for human health and safety that high standards of fish quality must be maintained at every stage of the food chain, from capture to consumption. Freshness and quality of fish at each step of the fish production chain can help manufacturers make safe, high-quality, and healthy fish meats, giving them an exceptional price in Global demand.

In spoilage, only a subset of these contaminants can colonize and multiply in large numbers. The spoilage association in aerobically preserved fish is generally composed of Gram-negative psychrotrophic non-fermenting rods. There are several bacterial species on the surfaces of fish. According to their development temperature range, all temperate water fish bacteria are classified as either psychrotrophs or psychrophiles. Psychrotrophs (cold-tolerant) bacteria can thrive at 0°C; their growth is most efficient around 25°C. Bacteria known as psychrophiles (cold-loving) thrive best at temperatures between 15°C and 20°C. The term "spoilage association" has been coined for such a microbial community, but the precise mechanism by which one bacterial group dominates another closely related group is not always fully understood. Thus, under aerobic iced storage, the flora is virtually entirely constituted of *Pseudomonas sp.* and *S. putrifaciens*. Gram-negative, fermentative bacteria (such as *Vibrionaceae*) are responsible for spoiling unpreserved fish. At room temperature (25°C), the microflora is dominated by mesophilic *Vibrionaceae*, especially if the fish are taken in contaminated waters. Fish spoilage is mainly caused by microbial growth, which creates flavour-altering amines, biogenic amines, organic acids, alcohols, aldehydes, and ketones. Internal fish tissue is often considered sterile, but the bacteria on the slime layer of the skin, gills, and gut would invade after the death of the fish. Factors such as high water activity and low acidity (pH > 6) of fish contribute to the rapid proliferation of microorganisms, which cause negative changes in the fish's appearance, texture, taste, and odour, diminishing its quality. Fish muscle consists of proteins, lipids, carbohydrates, water, and amino acid components, such as trimethylamine oxide (TMAO), urea, taurine, creatine, free amino acids,

and trace glucose. In addition to psychrotrophic, aerobic, and facultative anaerobic Gram-negative bacteria, such as *Pseudomonas*, *Moraxella*, *Acinetobacter*, *Shewanella putrefaciens*, *Vibrio*, *Flavobacterium*, *Photobacterium*, and *Aeromonas*, Gram-negative bacteria also contribute to fish spoilage.

Table 1. Specific spoilage Organisms in the fish and fishery products:

Storage temperature	Packaging atmosphere	Dominating microflora	Specific spoilage organisms (SSO)
0°C	Aerobic	Gram-negative psychrotrophic, non-fermentative rods (<i>Pseudomonas</i> spp., <i>S. putrefaciens</i> , <i>Moraxella</i> , <i>Acinetobacter</i>)	<i>S. putrefaciens</i> <i>Pseudomonas</i>
0°C	Vacuum	Gram-negative rods; psychrotrophic or with psychrophilic character (<i>S. putrefaciens</i> , <i>Photobacterium</i>)	<i>S. putrefaciens</i> <i>P. phosphoreum</i>
0°C	MAP	Gram-negative fermentative rods with psychrophilic character (<i>Photobacterium</i>) Gram-negative non-fermentative psychrotrophic rods (1-10% of flora; <i>Pseudomonas</i> , <i>S. putrefaciens</i>) Gram-positive rods (LAB 2)	<i>P. phosphoreum</i>
5°C	Aerobic	Gram-negative psychrotrophic rods (<i>Vibrionaceae</i> , <i>S. putrefaciens</i>)	<i>Aeromonas</i> spp. <i>S. putrefaciens</i>
5°C	Vacuum	Gram-negative psychrotrophic rods (<i>Vibrionaceae</i> , <i>S. putrefaciens</i>)	<i>Aeromonas</i> spp. <i>S. putrefaciens</i>
5°C	MAP	Gram-negative psychrotrophic rods (<i>Vibrionaceae</i>)	<i>Aeromonas</i> spp.
20-30°C	Aerobic	Gram-negative mesophilic fermentative rods (<i>Vibrionaceae</i> , <i>Enterobacteriaceae</i>)	Motile <i>Aeromonas</i> spp. (<i>A. hydrophila</i>)

(Courtesy: FAO fisheries technical paper – 348)

Microorganisms develop spoilage chemicals during the preservation of fresh fish. Bacterial proliferation results in the formation of a slime layer, the darkening of the gills and eyes (in whole fish), and the loss of muscle texture (softened due to proteolysis). The volatile molecules produced by protein putrefaction cause odours such as fishy (due to trimethylamine) and spoilage. Numerous proteolytic and hydrolytic enzymes are produced by *Pseudomonas*

putrificans, *Pseudomonas fluorescens*, and other spoilage bacteria when they proliferate and multiply fast. *Pseudomonas fluorescens* is responsible for fish's greenish-yellow hue, whereas *Micrococcus*, *Bacillus* and *Sarcina* are responsible for the yellow and red hues, respectively. Yeasts and moulds are responsible for the chocolate-brown hue, and *Streptomyces* for the musty stench.

4.0 Seafood borne pathogens:

In addition to human non-pathogenic bacteria species and the natural microflora of aquatic habitats, pathogenic bacteria are prevalent in fish. According to the European Food Safety Authority, *Campylobacter*, *Salmonella*, *Yersinia*, *E. coli*, and *Listeria monocytogenes* are responsible for significant foodborne outbreaks across the globe. However, not all bacteria are linked to outbreaks of foodborne illness caused by the eating of contaminated fish and fish products. Meanwhile, *L. monocytogenes*, *Vibrio spp.*, *Salmonella*, *Yersinia spp.*, and *C. botulinum* are particularly interested. These pathogens have a broad distribution in aquatic habitats and are associated with significant death rates in people due to illnesses such as listeriosis, botulism, and *V. vulnificus* infection. Thus, along with the nutritional advantages of consuming fish, there is also a possible danger to human health.

5.0 Factors contributing to the risk of seafood-borne diseases:

The increasing demand and consumption of seafood in various countries has led to a heightened susceptibility to bacterial and viral contamination in seafood products. The consumption of seafood may occur at various stages, including primary production, handling, transferring, and preparation. The incidence of disease related to contaminated seafood has significantly risen in the past decade. Consequently, there has been a global increase in awareness regarding seafood-related illnesses due to the growing threat they pose. Seafood-borne outbreaks can arise due to the presence of parasites, bacteria, and viruses. The resulting symptoms can range from mild gastroenteritis to severe, life-threatening infections. Shellfish can be susceptible to contamination by various viruses, including norovirus and hepatitis A virus, as well as bacteria such as *Vibrio spp.*, *Shigella spp.*, and *Salmonella spp.* Additionally, protozoan parasites like *Toxoplasma gondii*, *Cyclospora spp.*, and *Cryptosporidium spp.* have been identified as potential contaminants of shellfish. Additionally, both freshwater and marine environments can harbour various zoonotic pathogens, which can be found in finfish and cephalopods. There have been multiple instances of *Vibrio spp.* infection documented due to consuming shellfish, specifically oysters. *Salmonella* outbreaks are frequently linked to

sushi consumption, whereas contamination of smoked mussels, salmon, and other fish has been associated with outbreaks caused by *Listeria spp.* *Vibrio spp.* has been identified as the causative agent in numerous food-borne outbreaks. Disease transmission can occur through zoonotic bacteria, such as Salmonella, which can cause illness in aquatic species and humans. The potential for seafood contamination leading to illness can be classified as either high or low, although there may be differing opinions among authors regarding this categorization. Food products that are considered high risk include mollusks and shellfish, raw and lightly processed fish products, as well as fish products that undergo processing at low temperatures. Seafood options with a low risk factor encompass smoked dried fish, semi-preserved fish, fresh or frozen fish and crustaceans, as well as heat-treated fish that is canned. The consumption of dry and heavily salted fish presents minimal risk of infection or pathogen transmission. Despite the implementation of the Hazard Analysis Critical Control Point (HACCP) system aimed at mitigating seafood-borne diseases, the prevalence of contaminated seafood remains a significant contributor to food-borne infections in the United States. Interestingly, it has been observed that seafood-borne outbreaks are frequently associated with intoxication rather than infection. This can be attributed to the significant number of reported cases of histamine food poisoning. The presence of naturally occurring pathogens such as *Vibrio spp.* and *Aeromonas spp.* in sea water and sediment can pose a significant risk to consumers who consume seafood. This environmental contamination should be considered as an important factor to be addressed. Additionally, there is a potential for inter cross-contamination between different operations.

6.0 Methods for reducing microbial load:

Physical damage, such as scale loss, bruising, and gut bursting, increases the number of sites available for bacterial attack and spread. Furthermore, cortisol levels rise during prolonged stress, affecting fillet quality. After the catch, fish may be held in the vessel for a few hours or weeks in melting ice, cooled brine, or -2 °C saltwater. Inadequate circulation of chilled brines may lead to the localized anaerobic development of specific microbes and spoiling, accompanied by the formation of off-odours. Used refrigerated brines may be polluted with many psychrotrophic spoiling bacteria, and reusing them can enhance the cross-contamination of other fish with these microbes. Increasingly, and mainly when fish is held on board for extended durations, freezing facilities (-18°C) may be employed to preserve the harvest (if possible). Fish may be eviscerated before marine storage, which has pros and cons. Intestinal enzymes and gut bacteria may discolour, degrade, and off-flavour un-eviscerated fish. In eviscerated fish, the incisions reveal microbial-vulnerable flesh. When eviscerating at sea,

remove all stomach contents and wash the corpse before refrigerating, icing, or freezing. Whether to gut the catch at sea depends on its size.

7.0 Practices to control the pathogenic bacteria:

There are several bacterial pathogens that pose a microbiological hazard to seafood, including *Vibrio spp.*, *Salmonella spp.*, *L. monocytogenes*, *S. aureus*, *C. botulinum*, *Shigella spp.* and *Aeromonas spp.* These bacteria have the potential to contaminate seafood products at any stage of the supply chain, from farm to table. It is imperative to implement effective strategies for the control and prevention of bacterial hazards in the fish industry. To minimise bacterial hazards, it is important to maintain the microbiological water quality of domestic capture, practise proper post-harvest care, adhere to good manufacturing practises (GMP), follow good hygienic practises (GHP), and implement HACCP protocols. Foodborne intoxications can be effectively managed through the implementation of appropriate refrigeration practises for seafood and the consistent monitoring of the cold chain throughout the entire production process until consumption. Additional measures to mitigate the occurrence of food-borne outbreaks resulting from the consumption of seafood include educating consumers on appropriate food handling practises, ensuring proper preparation techniques, and implementing effective seafood storage methods. Maintaining regular surveillance is crucial for evaluating the efficacy of both present and future prevention strategies.

Suggested Readings:

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- Ryder, J., Iddya, K. and Ababouch, L., 2014. Assessment and management of seafood safety and quality: current practices and emerging issues. *FAO fisheries and aquaculture technical paper*, (574), p.I.