Recent advances in preservation and freshness monitoring methods for fish: a review on the quality and structural changes of fish after slaughter

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Abstract. Fish is a popular and nutritious food, which faces rapid spoilage due to its high protein content, leading to a short shelf life. Consequently, research has been directed towards effective preservation techniques and monitoring methods to address this challenge. This review delves into advancements in fish preservation and freshness monitoring, highlighting the perishable nature of fish owing to its protein content. It examines post-slaughter quality and structural changes in fish meat, recent preservation methods including physical methods and packaging approaches, and proposes a theoretical framework for further development in fish preservation and freshness assessment. Key areas of focus include sensory quality changes, pH value, water-holding capacity, thiobarbituric acid reactive substance value, TVB-N value, structural changes post-slaughter, stress-related muscle metabolism alterations, enzymatic myofibrillar proteolysis, and lipid impacts on fish quality. Additionally, the review discusses preservation methods like physical treatments, ultrasound, modified atmosphere packaging, vacuum packaging, and active packaging, along with monitoring techniques such as sensory bionic technology and spectroscopy technology for assessing fish freshness.

Keywords: fish preservation; freshness monitoring; structure change; intelligent packaging technology.

1. Introduction

Fish flesh stands as a pivotal nutritional cornerstone in human diets, boasting a rich array of nutrients and commendable nutritional value. Notably, fish are abundant in essential compounds such as taurine, choline, NAD, and phosphatidylcholine, pivotal for maintaining human health[1]. DHA, a prominent type of omega-3 fat prevalent in fish, constitutes approximately 60% of the brain's fats, underlining its critical role in promoting healthy brain development, particularly during fetal development and early childhood, as emphasized by Bord Bia - The Irish Food Board.

Despite its nutritional prowess, the perishable nature of fish poses a considerable challenge in preserving its quality post-harvest. Fresh fish, heralded as a primary source of animal protein, is susceptible to various forms of deterioration stemming from slaughtering activities and subsequent handling. The intricate interplay of chemical, physical, and microbial processes occurring throughout the supply chain stages, including processing, storage, and transport, exacerbates the susceptibility of fish meat to spoilage[2]. High moisture content, protein, fat, weak muscle tissue, unhygienic handling practices, and ambient temperature fluctuations collectively accelerate the spoilage process. The onset of deterioration in fish meat post-mortem initiates a multifaceted cascade of events, characterized by protein degradation, oxidative rancidity, and microbial activity. Sensory assessments, alongside measurements of fish pH, TVB-N levels, and microbial counts, emerge as pivotal indicators influencing the maintenance of freshness during storage.

This review focuses on quality and structural changes in fish after slaughter, as well as recent advances in fish preservation and freshness monitoring. Studies have pointed out that the quality and structure of fish meat can be affected by slaughtering activities, while fresh meat quality is susceptible to chemical and physical changes or microbial activities. In addition, recent research on fish preservation methods, including physical methods and various packaging methods, was explored.

Finally, a theoretical framework for further development of fish preservation and freshness assessment is proposed (Figure 1).

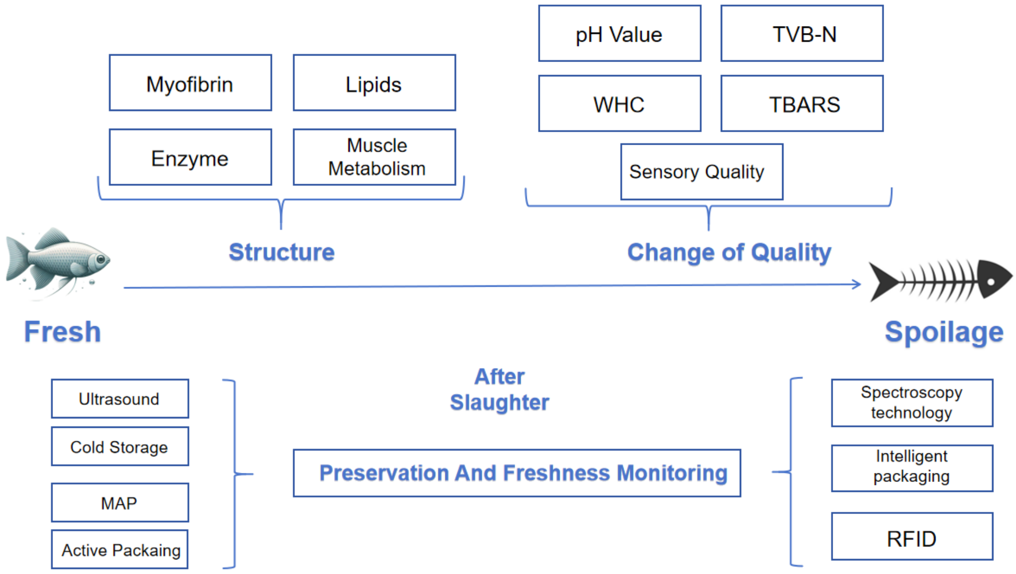

Figure 1. Graphical summary of fish freshness assessment, monitoring, and preservation methods

2. The quality of the fish changes after the slaughter

2.1. Changes in sensory quality

Sensory evaluation, particularly through attributes like appearance, smell, texture, and taste, plays a crucial role in assessing fish quality. Quality Index Method (QIM) evaluates key characteristics such as eyes, gills, skin color, and odor to provide a comprehensive quality score, though its development faces challenges such as subjectivity and complexity[3].

2.2. pH value

After slaughter, fish undergoes rigor mortis, maturation, autolysis, and spoilage, affecting meat quality and pH value changes[4, 5]. The pH variation influences meat properties such as color and tenderness, with alkaline substances (TVB-N, TMA, DMA and ammonia accumulate) accumulating during degradation, leading to pH increase. This increase reflects fish flesh freshness to some extent[6, 7].

2.3. Water-holding capacity and drip loss

Water-holding capacity (WHC) impacts taste and juiciness, with poor WHC resulting in a rough taste and low yield[8]. Increased drip loss during storage indicates reduced WHC, exacerbated by protein degradation. Studies have shown that the moisture content of fish fillet decreases with the increase of storage time and the drip loss increases continuously. During the late storage period, fillets showed severe protein degradation and denaturation, which reduced WHC and increased the drip loss and other parts during cooking. Vacuum packaging and ice temperature storage mitigate these effects, extending shelf life[9] .

2.4. Thiobarbituric acid reactive substance value

Thiobarbituric Acid Reactive Substance (TBARS) assays detect lipid oxidation, a major factor in meat spoilage. Previous studies have shown that fillet is unacceptable and inedible when the TBA value exceeds 1mg MDA/kg. Fat oxidation leads to ranity of meat, producing unpleasant smells, which will also reduce the nutritional value of meat. Fat oxidation produces aldoketones such as MDA, which reacts with TBA to produce colored substances[10]. Elevated TBARS values indicate unacceptable rancidity levels, mitigated by oxygen sequestration and lower storage temperatures [9].

2.5. TVB-N value

TVB-N content is one of the critical safety evaluation indexes in seafood, a better indicator of their suitability and freshness than TBA for high-protein and low-fat aquatic products[9-11]. TVB-N content reflects fish freshness, increasing due to protein decomposition, which produces ammonia, amines, indole, hydrogen sulfide and other substances, accompanied by putrid odor[12, 13]. Alkaline nitrogen-containing substances produced by protein decomposition continue to accumulate, showing a continuous rise in TVB-N value. EU regulations consider fish with TVB-N levels exceeding 35 mg/100 g as spoiled. However, understanding its relationship with other freshness parameters remains limited[7, 12].

3. Structural changes of the fish after slaughter

After slaughter, fish undergoes a series of desirable biochemical changes, including acidification, muscle protein gelation, and lipid degradation, which influence its texture and structure[14]. Dark-fleshed species, rich in lipids and sarcoplasmic proteins, experience altered elasticity and cohesion due to protein gelation, reflecting freshness quality[14, 15].

3.1. Stress-related changes in post-mortem muscle metabolism

Post-mortem muscle metabolism undergoes stress-related changes, with anaerobic glycolysis persisting but glycogen reserves depleted, inhibiting glycolytic enzymes due to lactic acid accumulation. ATP depletion post-mortem leads to muscle contraction, influencing meat quality traits such as color and texture. Rapid pH decrease correlates with paler meat color and reduced water-holding capacity[16].

3.2. Myofibrin

Myofibrillar protein solutions from fish exhibit decreased hydrophobicity and $Ca2^{+}$-ATPase activity over time during 4°C storage, with alterations in protein structure observed through texture analysis. Increased size and fractal dimension of protein particles are observed on the 3rd day post-slaughter[17].

3.3. Enzymes in myofibrillar proteolysis

Enzymes such as calpain and cathepsins play significant roles in post-mortem muscle breakdown, cleaving proteins and softening fish flesh. The calpain protease system targets hydrophobic amino acids, while lysosomal cathepsins contribute to protein degradation, leading to textural changes in fish flesh[18].

3.4. Lipids

Fish lipids undergo oxidation and hydrolysis post-slaughter, impacting its quality. Fatty fish, rich in polyunsaturated fatty acids, are highly susceptible to oxidation, leading to the formation of taste-altering compounds. Enzymatic and nonenzymatic reactions catalyze lipid degradation, with metal ions and lipoxygenase playing key roles. Cooking and freezing/thawing can mitigate enzymatic activity.

4. Fish preservation methods

4.1. Physical treatment

4.1.1. Storage conditions

Reduced temperature storage has limited effectiveness due to the inherent characteristics of fish, such as high unsaturated fatty acid content and unstable proteins, making freezing at -30°C a common preservation method[19]. Freezing inhibits microbial growth and enzyme activity, extending shelf life, although ice crystal formation can impact texture upon thawing[20-22]. Emerging thawing methods, including pulsed electric fields, high pressure, and ultrasound, aim to mitigate structural damage and maintain quality[23]. The study found that slurry ice improved quality stability and shelf-life extension[24].

4.1.2. Ultrasound

Ultrasound, with its directional, high-energy, and cavitation effects, has gained traction in food processing, including fish preservation[25]. By inducing structural changes in proteins, ultrasound enhances meat quality and shelf life, making it a promising technology in the food industry[26-29].

Studies reveal that ultrasound-induced cavitation alters protein spatial conformation and functional properties, impacting covalent and non-covalent interactions[30-31]. Additionally, ultrasound-assisted phenolic covalent modification improves thermal stability and antioxidant capacity[32-34].

Various combinations with ultrasound show enhanced effects in food processing, such as ultrasound with carvacrol effectively controlling Bacillus cereus[35]. Combining slightly acidic electrolyzed water (SAEW) with ultrasound enhances bactericidal effects in tuna fish, while cinnamon essential oil (CEO) integrated into nanofiber mats demonstrates antimicrobial properties[36, 37]. Moreover, SAEW combined with ultraviolet light enhances antibacterial effects on Staphylococcus aureus, further potentiated with ultrasound assistance[38]. Multifrequency ultrasound-assisted thawing maintains myofibril stability and reduces lipid oxidation in frozen large yellow croaker.

4.2. Packaging

4.2.1. Modified Atmosphere Packaging and Vacuum packaging

Modified Atmosphere Packaging (MAP) technology alters the gaseous environment surrounding food to extend shelf life and minimize quality defects[33]. By adjusting gas compositions, MAP effectively retards deterioration, preserving food freshness for extended periods. Vacuum packaging, a variant of MAP, removes atmospheric gases to inhibit oxidative reactions and microbial growth, proving effective in extending shelf life[39]. Shelf-life tests done by P. Yesudhason et al. determined optimal gas composition and sodium acetate concentrations in MAP. This combined approach (MAP with 70% CO2:30% O2 and 1% sodium acetate) proved effective in significantly extending the shelf life of raw fish products[40]. In addition, Zhijun Yang et al. demonstrated MAP effectively inhibited microbial growth, maintained water-holding capacity (WHC), and protected against lipid and protein oxidation[41].

4.2.2. Active packaging

Active packaging integrates substances like antioxidants and antibacterial agents into packaging materials to maintain food quality and safety.

4.2.2.1 Antioxidation

Electrospinning is a versatile technique for producing nonwoven fiber mats at a sub-micron or nano-scale using natural or synthetic polymers. These mats offer advantages such as larger specific surface area, high porosity, and responsiveness to environmental changes. Carvacrol(5-isopropyl-2-methylphenol, CA), a key component of thyme and oregano oils, is often used in food packaging due to its strong antibacterial and antioxidant properties[42-44]. Min Guo et al. developed a dual-layer fiber mat using electrospinning, with one layer containing pH-responsive purple sweet potato extract and the other layer incorporating antibacterial carvacrol. This innovative packaging material shows potential for enhancing food safety and preservation by monitoring color changes in response to pH variations[45].

4.2.2.2 Antibacterial

Lauroyl Arginate Ethyl (LAE) is a relatively new antimicrobial substance used in food preservation. It belongs to cationic surfactants and can be broken down into ethanol, arginine, and lauric acid, which are safe for human consumption. Previous studies have explored incorporating LAE into packaging films made of materials like poly(lactic acid), polyvinyl alcohol, chitosan, and gelatin[46-47]. Dong et al. found that electrospun fish gelatin films containing LAE could extend the refrigerated shelf life of large yellow croaker fillets by about three days, showing potential as an antibacterial method for fish preservation. Carvacrol (CA) has broad-spectrum antimicrobial properties against gram-positive bacteria, gram-negative bacteria, and fungi, as well as anticancer and anti-inflammatory effects. Dai et al. used covalent organic frameworks (COFs) to immobilize silver nanoparticles in a chitosan film, which effectively controlled pH increases during the storage of white crucian carp, thereby reducing microbial growth within the fish[48].

5. Monitoring methods for freshness of fish and meat

Quality assessment of fish freshness typically encompasses early autolysis and later microbial proliferation stages. Autolysis, primarily driven by tissue enzymes, involves the degradation of adenosine triphosphate (ATP) to inosine (Hx) via adenosine diphosphate (ADP), adenosine monophosphate (AMP), and inosine 5'-phosphate (IMP). Protein breakdown yields various amines, including total volatile nitrogen (TVB-N) and biogenic amines (BA), while oxidation of fatty acids results in aldehydes and ketones. Additionally, glycogen breakdown produces lactate. Furthermore, specific deteriorating characteristics manifest in the color of different body parts and the emission of multiple odors simultaneously. Traditional monitoring techniques focus on sensory, chemical, physical, and microbial attributes, but recent advancements offer accurate, rapid, cost-effective, and noninvasive alternatives[49].

5.1. Sensory bionic technology

Organoleptic quality serves as a vital indicator in assessing fish freshness, with the Quality Index Method (QIM) being a primary approach. QIM assesses appearance, texture, smell, and color, providing initial insights into fish freshness. Fresh fish typically exhibits bright eyes, smooth skin, elastic flesh, and a pleasant odor, whereas spoiled fish presents cloudy eyes, thick skin, soft flesh, and unpleasant smells. Despite its significance, sensory evaluation suffers from subjectivity and lack of quantifiability[50].

5.2. Spectroscopy technology

Fluorescence spectroscopy, Fourier Transform Infrared Spectroscopy (FTIR), Raman spectroscopy, and Hyperspectral imaging (HSI) offer non-destructive means to evaluate fish and meat quality. Fluorescence spectroscopy identifies various meat compounds and assesses TVB-N levels[39]. FTIR and Raman spectroscopy, with their sensitivity and real-time analysis capabilities, are promising[51]. NIR spectroscopy intelligently assesses histamine levels in fish, while HSI captures both physical and chemical information, distinguishing fresh from thawed products[52]. However, challenges include high costs and slow data processing[53]. Omwang et al. also found the potential of fluorescence technology in non-destructive and rapid freshness assessment of fish[54].

5.3. Intelligent packaging

Intelligent packaging employs indicators, sensors, and Radio Frequency Identification (RFID) tags for real-time monitoring of fish freshness. Indicators, primarily pH-sensitive dyes, undergo color changes in the presence of TVB-N. Sensors measure various indicators like temperature and odor, while RFID tags enable real-time monitoring of fish parameters. These technologies offer crucial support in maintaining fish freshness and safety. Khumngern et al. introduce R-Paper, a smartphone-based histamine sensor, offering semi-quantitative results and successful detection of hyaluronic acid in fish samples[55]. Gopalakrishnan et al. develop a sensor tag for selective monitoring of hypoxanthine as a spoilage biomarker, distinguishing between spoiled and fresh products[56]. Chaithra K. P. et al. propose "R-Paper" as a user-friendly indicator for cost-effective fish freshness monitoring, highlighting its potential for widespread use. These advancements leverage smartphone applications, selective sensors, and simple indicators for effective fish quality assessment.

5.4. Radio Frequency Identification

RFID technology facilitates automated target identification and data reading, integrating gas sensors for real-time freshness monitoring, providing comprehensive, highly sensitive, accurate, and rapid analysis.The RFID sensor's equivalent circuit forms an LCR circuit, interacting with the pickup coil and the card reader to read sensor information and monitor impedance changes. PANI polymer in the sensing film reacts with TVB-N, altering film conductivity. Although effective, concerns about costs limit its applicability to high-value fish products[49].

6. Conclusions and future research directions

This review underscores the challenges in preserving fish freshness and explores its complex spoilage process， which will provides a basis for the development of new preservation technologies for fish and meat. It emphasizes the role of sensory evaluation, pH changes, water-holding capacity, and the drip loss as freshness indicators. And the preservation methods of fish are emphasized. The paper concludes by highlighting the use of intelligent packaging technology.

In the future, fish freshness assessment will focus on innovative, non-invasive methods such as sensory bionic technology and smartphone-based colorimetric sensors. Understanding external factors like temperature and packaging materials is crucial for freshness management. Research will explore post-mortem changes, optimizing ultrasound technology for enhanced quality improvement. Additionally, studies will investigate antimicrobial agents and modified atmosphere packaging to inhibit microbial growth, extend shelf life, and assess their impact on fish product quality and moisture.

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