Packaging Interventions in Low Temperature Preservation of Fish- A Review

Abstract
Consumers demand for fresh or fresh like fish products without altering its natural quality attributes will be met by using low temperature preservation particularly chilling. Low temperature preservation is widely practiced in the industry to overcome the spoilage of fish. However, low temperature alone has limited shelf-life which lead to the introduction of innovative packaging interventions ranging from high barrier packaging material to most recent active packaging technologies. Vacuum packaging (VP) and modified atmosphere packaging (MAP) helped in reviving the fresh fish industry as they helped in improving safety as well as shelf life considerably. The recent active packaging technologies especially O2 scavenger and CO2 emitters have additional advantage of very limited capital investment compared to VP and MAP technologies. Present article gives a brief of recent advancements of these packaging innovations for fish preservation under chilled or refrigerated storage condition.

Keywords: Fish; Chilling; Vacuum packaging; Modified atmosphere packaging; Active packaging

Abbreviations: VP: Vacuum Packaging; MAP: Modified Atmosphere Packaging; PUFA: Poly-Unsaturated Fatty Acids; EPA: Eicosapentaenoic Acid; DHA: Docosahexaenoic Acid; NPN: Non-Protein Nitrogen; GRAS: Generally Recognized-As-Safe; MA: Modified Atmosphere; AP: Active Packaging

Highlights
a. Chilling / refrigerated fish preservation is the most widely preferred as it has limited effects on sensorial quality.

b. Vacuum packaging and modified atmosphere packaging improves safety and shelf life of fishes.

c. Active packaging technologies are most effective packaging interventions in improving shelf life of fish.

Introduction
Consumption of fish is increasing globally due to increasing evidence of positive benefits for medical conditions like constipation, cardiovascular diseases, obesity, hypertension and certain variety of cancer. World per capita apparent fish consumption increased from an average of 9.9 kg in the 1960s to 19.2 kg in 2012 [1]. Fish is the main source of protein in many parts of the world. Fish is a rich source of dietary proteins and omega-3 (n-3) fatty acids which reduces cholesterol levels. Various health benefits have been attributed to the consumption of fish like cardioprotective benefits, improved pregnancy outcomes, fewer preterm and low birth weight deliveries. Though fish is highly nutritious, it is also highly perishable due to the presence of high moisture, protein and highly oxidizable poly-unsaturated fatty acids (PUFA), making it vulnerable to various biochemical, physical and microbial forms of deterioration throughout the production chain. Proper preservation of fish assumes greater importance to reduce the wastage of nutritious food commodity. Among the various preservation methods available, low temperature preservation especially chilling or refrigerated storage has attracted interest of many researchers as it induces minimal changes in the texture and other characteristics of fish. However, it has limited shelf life depending on the species of fish. Although various researchers have reported beneficial effects of using innovative packaging technologies like vacuum packaging, modified atmosphere packaging and active packaging technologies on the safety and shelf life of fish products, there is no report on the detailed review in these aspects. In this review, an attempt was made to compile data obtained from literature related to the packaging interventions to enhance the quality and shelf life of fish products preserved in low temperature storage conditions.

Nutritive benefits of fish
Health, nutrition and convenience are the most demanding, challenging and major drivers in the global food industry. The fresh fish and seafood’s rank third (12% growth rate) among the food categories with the fastest overall growth worldwide in 2006, next to drinkable yogurt (18%) and fresh soup (18%) [2]. Seafood products have attracted considerable attention as a source of high amounts of important nutritional components like high-quality protein, essential vitamins and minerals and healthful polyunsaturated fatty acids to the human diet [3-5]. Fish is regarded as one of the cheapest animal protein sources and it accounts for about 40% of the total animal protein intake of an average person in the tropics [6]. Societies with high fish intake
have considerably lower rates of acute myocardial infarctions, other heart diseases and atherosclerosis, arthritis, asthma and other inflammatory or self-immune illnesses or certain types of cancer [7-10]. The highest life expectancies in Japan [2] could be attributed to their high seafood consumption. The above mentioned benefits are thought to be due to rich omega-3 polyunsaturated fatty acids content (Ω-3 PUFA). Furthermore, dietary intake of Ω-3 PUFA was inversely related to the risk of impaired cognitive function. Ω-3 PUFAs are also critical for normal neural and visual development in the human foetus. The two important Ω-3 PUFAs, EPA (Eicosapentaenoic acid) and DHA (Docosahexaenoic acid) are available to consumers mainly through a diet rich in fish. Studies also indicated that dietary factors such as DHA have neuroprotective effects which help in preventing Alzheimer’s disease [11]. Nutrition recommendations in many countries suggest that fish should be an integral part of a nutritious human diet. In many countries, seafood consumption has increased considerably due to the above benefits. As fish is highly nutritious it is also highly perishable due to its high moisture content, protein and highly oxidizable PUFA. Due to this fish and fish products are vulnerable to various biochemical, physical and microbial forms of deterioration throughout the production chain (catch to retail sale) which leads to the breakdown of protein and lipid fractions and the formation of amines (volatile and biogenic) and hypoxanthine. In spite of the potential health benefits related to fish consumption, eating of fish containing oxidized fatty acids limited health benefit. The fish lipid containing high amount of polyunsaturated fatty acids are known to be highly susceptible to oxidative breakdown [12-15] and heat catalysts strongly for the initiation of lipid peroxidation [16-20]. These oxidation products can lead to certain medical disorders, such as higher risk of atherosclerosis [21], oxidative stress, and exacerbate at heterogenesis by incorporating into lipoproteins [22], and serious alterations in membrane composition, fluidity, and function [23]. These leads to deterioration in sensory quality, a loss of nutritional value, and negative modifications of the physical properties of fish muscle [24-26]. In order to increase the average amount of fish consumption, good quality seafood that is well prepared and conveniently packed should be available [27].

Spoilage of Fish

The principal constituents of fish muscle are water (66-89%), protein (16-21%), lipids (0.25-25%) and ash (1.2-1.5%) [28]. The myofibrillar protein, actin and myosin (2/3 of proteins) play an essential role when fish swims. The water soluble proteins are mostly enzymes [28]. Fish and shellfish are highly perishable, because of the intrinsic factors (for example, high $a_w$ neutral pH, presence of autolytic enzymes, highly digestible proteins and highly unsaturated fatty acids, large amounts of non-protein nitrogen (NPN), the presence of TMAO as part of NPN fraction, mainly in marine fishes) and the extrinsic factors (for example, temperature, processing and packaging atmosphere) [29-36]. The fish spoilage is also due to the high metabolic rate of the species, with enzymes that remain active after death, causing protein hydrolysis [37]. The rate of deterioration is associated with many factors such as fish species, size and lipid content, condition at the moment of capture, microbial load, and storage temperature [38-40]. Enzymatic and chemical reactions are usually responsible for the initial loss of freshness, whereas microbial activity is responsible for the obvious spoilage, thereby establishing product shelf life and resulting in heavy economic loss. In living muscle cells, a wide variety of controlled biological (enzymatic) reactions occurs. In the dead cells, these reactions continue or take place in a disturbed way, and several other biochemical and enzymatic reactions are triggered in fish muscle, resulting in the gradual loss of fish freshness [37,41]. Animals maintain energy for living through the oxidation of all kinds of organic compounds in their bodies. Due to the decrease in oxygen supply to the tissue after fish death, anaerobic metabolism takes over, resulting in the conversion of glycogen to lactic acid. With a decrease in ATP content and creatinine phosphate [42,43], actin and myosin gradually associate to form inextensible actomyosin (onset of rigor mortis) which leads to stiffening and rigid condition of muscle tissues. Fish generally exhibit rigor mortis starting from about 1 to 6 h after death when ATP is being depleted below a certain critical level in the muscle [44]. The reduced pH due to accumulation of lactic acid and lowering ATP interact with other biochemical processes occurring after death, especially myofibrillar proteolysis. Lysosomal cathepsins, neutral calcium-activated cathepsins and proteasomes are the three proteolytic systems which should bring about the post mortem changes resulting in flesh deterioration. Calpains may initiate the disintegration of the Z line by a titin cleavage [45], which weakens the titin/α-actinin interaction and results in the release of intact α-actinin from Z lines [46]. Similarly, cathepsins and proteasomes have been reported to induce muscle softening [47,49]. Rigor mortis disappears in a short time even under the most favourable storage conditions. Immediately thereafter, the decomposition of the highly complex protein of the fish muscle into simpler protein, polypeptides and amino acids starts to take place. These changes are known as autolysis. While autolysis is proceeding, bacterial decomposition begins, which is the most complex and important of all the changing processes. Although the flesh of the freshly caught fish is sterile, the external surfaces carry a considerable bacterial load. In the gutted fish, the bacteria proceeds from the kidney, along the cardinal vein, which lies beneath the backbone in the caudal region of the fish and breaking up the corpuscles and finally entering the tail flesh. Apart from this, improper handling also causes the bacterial contamination. A bacterium thus entered degrades fish constituents, particularly non-protein nitrogen compounds and induces the development of off-odors and flavors typically associated with fish spoilage [50]. The rate of spoilage depends on several factors, the nature of fish species and the handling and storage conditions etc. [26,51]. These changes coupled with the chemical activities mainly lipid oxidation, leads to loss of quality and subsequent spoilage which makes the fish unfit for human consumption. To minimize these changes there by spoilage and to maintain the quality of fish products for prolonged period various preservation methods are being employed worldwide.

Fish Preservation

The two major problems with respect to marketing and distribution of seafood’s are their high perish ability and poor hygienic quality [26]. To overcome these problems various preservation methods are being practiced. The principle aim
of fish preservation is to delay, reduce or inhibit the enzymatic, chemical and microbial spoilage. This is achieved by controlling the storage temperature, maintaining proper water activity, pH or by using chemical preservatives [52]. The concept of temperature control as a means to prolong shelf life is well understood, and the response of enzymatic, chemical and bacterial activity to temperature has been widely studied [53-57]. Fish spoilage depends mostly on temperature, which controls to a large extent the bacterial and the autolytic breakdowns. Preservation technique by temperature control is of two ways, either by lowering or increasing the temperature. Methods involving the lowering of temperature are chilling and freezing. In chilling, the temperature of the fish is lowered immediately to reduce the autolytic and bacterial changes. To slow down the mechanisms involved in quality loss, the fish should be refrigerated immediately after capture until consumed [31,58]. In freezing, the products are cooled below a temperature of -35°C to arrest the enzymatic and bacterial activities completely to prolong the shelf life [59]. The freezing and frozen storage of fish have been largely used to retain their sensory and nutritional properties before consumption [60-62]. However, protein denaturation, toughening of the texture, excessive drip loss during thawing and high operating costs associated with freezing and frozen storage are some of the disadvantages [63,64]. Canning is another method, in which the temperature is increased to more than 100°C to inactivate all the enzymes and bacterial including spores [65-72]. Apart from these various other methods such as drying and salting, smoking, irradiation by gamma radiation, addition of chemical preservative agents, high pressure processing, sous-vide, pulsed electric field are employed as a means of preserving fish and fish products and have shown some degrees of effectiveness [73-78]. Each preservation method possesses some significant disadvantages like deterioration of fish quality and modification of freshness, lack of stability, and excessive cost [5]. However global demand for fresh mildly preserved, convenient with better keeping quality fishery products is increasing. This can be achieved by chilled storage of fish.

Chilling of fish

Shelf life extension of fish is of importance to allow the transport of products to distant markets at lower cost. Chilling has proven to effectively delay bacterial growth and prolong the shelf life of fish [79,80]. Various types of chilling systems have been used for seafood products including the conventional flake ice [25], refrigerated seawater [81], ozonised refrigerated water [82,83], slurry ice [84-87] and dry ice (solid carbon dioxide) [88]. Although chilling is very effective in delaying the spoilage it will not inhibit the spoilage completely as the enzymes and bacteria will be active at the chilled temperature. During chilled storage of fish, significant deterioration of sensory quality and loss of nutritional value have been detected as a result of changes in chemical constituents, that lead to a strong effect on the commercial value [5,25,51]. The storage life of chilled fish in flake ice, slurry ice, ozone-slurry ice of fish products are limited to 2-18 days depending on the species [89-97]. With the aim of reducing loss in freshness and improving the keeping quality of fish, chilling in combination with chemical preservative treatments, or innovative packaging techniques like modified atmosphere packaging, vacuum packaging and very recently active packaging techniques are employed.

Chemical treatments

The flesh of the freshly caught fish is regarded as sterile and their surfaces will carry contaminants, which are easily transferred to the flesh. The fresh and minimally processed fish products provide a good substrate for microbial growth. Such substrate may allow proliferation of human pathogenic organisms. Ensuring the safety of food products depends on minimizing the initial contamination with pathogenic microorganisms and inhibiting their development during handling and storage. Washing is one of the first processing operations to which a fish and shellfishes are subjected to reduce the contamination. Wash water chlorinated upto 2 ppm is routinely applied to reduce microbial contamination in produce processing lines. However, the use of chlorine is of concern due to the potential formation of harmful by-products [98] and can only achieve approximately 2 to 3 log reductions of native microflora [99]. Thus, there is much interest in developing safer and more effective sanitizers and preservatives. Various chemicals dip treatment such as Cetylpyridinium chloride [100], chlorine dioxide [101,102], or potassium sorbate [100], sodium lactate and lactic acid [103] Sodium and potassium lactates [104-106], sodium diacetate [107,108], sodium chloride (NaCl) and sodium nitrite (NaNO2) or potassium nitrate (KNO3) [109,110]. Sodium acetate, citrate and lactate [111,112,113] have been used in various food systems including fish products. Among these sodium salts of the low molecular weight organic acids, such as acetic, lactic and citric, have been used extensively to control microbial growth, improve sensory attributes and extend the shelf life of various food systems [114-116]. In addition to their suppressing effect on the growth of food spoilage bacteria by reducing the water activity, organic salts of sodium acetate, lactate and citrate were shown to possess antibacterial activities against various food-borne pathogens [117-119]. These salts are widely available, economical and generally recognized-as-safe (GRAS) by the U.S. Code of Federal Regulations, 21 CFR. Chemical preservative treatments coupled with the advanced packaging technologies and refrigerated storage will enhance the storage life of fish products.

Packaging innovations for fish preservation

Packaging makes food more convenient and gives the food greater safety assurance from microorganisms, biological and chemical changes so that the packaged foods may have a longer shelf life. Due to increased demands for greater stringency in relation to hygiene and safety issues associated with fresh and processed fish products, coupled with ever-increasing demands by retailers for cost-effective extensions to product shelf-lives and the requirement to meet consumer expectations in relation to convenience and quality (increased product range, easy use and minimum product preparation, provision of more product information and packaging impact on the environment), the food packaging industry has rapidly developed to meet and satisfy expectations. Packaging fresh fishery product is carried out to avoid contamination, delay spoilage, permit some enzymatic activity to improve tenderness, reduce weight loss, and where applicable, to ensure an oxymyoglobin or cherry-red colour in red meats at retail or customer level [120]. As a result, packaging has become an indispensable element in the food manufacturing
process. Among the packaging technologies developed by and for the food industry, vacuum and modified atmosphere packaging has led the evolution of fresh and minimally processed food preservation, especially in meat and fishery products for the past two decades.

Vacuum packaging (VP): It is well established that, the spoilage of fish is mainly due to the growth and proliferation of aerobic spoilage bacterial and the oxidation of lipid and pigments [121,122]. This can be reduced by using the vacuum packaging technique in which, the air present in the pack is completely evacuated by applying vacuum. Vacuum packaging is one of the natural preservation packaging methods which can greatly enhance the shelf life and overall quality of muscle foods for a long time [123]. In addition, packaging conditions that reduce the amount of oxygen present in the package are known to extend the shelf life of product by inhibiting the growth of aerobic spoilage bacteria [124]. This technique is particularly useful in fatty fishes [125], where the development of undesirable odour due to the oxidation of fat is the major problem. Ozogul F et al. [126] studied vacuum packaging fillets from sardines (Sardine pilchardus). Chouliara I et al. [127] reported the beneficial effects of combining vacuum packaging with irradiation in Sea breams. However, this technique is not suitable for all kinds of products. By employing this technique, it is not possible to remove the oxygen entrapped in the food. Moreover, the O₂ permeability of the packaging film will alter the package atmosphere. Hence more efficient technique has to be developed.

Modified atmosphere packaging (MAP): Marketing of modified atmosphere (MA) packaged foods have increased, as food manufacturers have attempted to meet consumer demands for fresh, refrigerated foods with extended shelf-life. It is also used widely, as a supplement to ice or refrigeration to delay spoilage and extend the shelf life of fresh fishery products while maintaining a high-quality end product. A modified atmosphere can be defined as one that is created by altering the normal composition of air (78% nitrogen, 21% oxygen, 0.03% carbon dioxide and traces of noble gases) to provide an optimum atmosphere for increasing the storage length and quality of food/produce [128,129]. Oxygen, CO₂ and N₂ are most often used in MAP [129,130]. Other gases such as, nitrous and nitric oxides, sulphur dioxide, ethylene, chlorine [129], as well as ozone and propylene oxide [130] have been suggested for a variety of products and investigated experimentally. However, due to safety, regulatory and cost considerations, they have not been applied commercially. These gases are combined in three ways for use in modified atmospheres: inert blanketing using N₂, semi-reactive blanketing using CO₂:N₂ or O₂:CO₂:N₂ or fully reactive blanketing using CO₂ or O₂:CO₂ [128,130].

The principle of MAP is the replacement of air in the package with a different fixed gas mixture. CO₂ is the most important gas used in MAP of fish, because of its bacteriostatic and fungistic properties [131]. It inhibits growth of many spoilage bacteria and the inhibition is increased with increased CO₂-concentration in the atmosphere and reduced temperature. CO₂ is highly soluble in water and fat, and the solubility increases greatly with decreased temperature. The solubility in water at 0°C and 1 atmosphere is 3.38g CO₂/kg water, however, at 20°C the solubility is reduced to 1.73g CO₂/kg water [132]. Therefore, the effectiveness of the gas is always conditioned by the storage temperature with increased inhibition of bacterial growth as temperature is decreased [133-135]. The solubility of CO₂ leads to dissolved CO₂ in the food product [132], according to the following equation:

\[ CO_2(g) + H_2O(l) \rightleftharpoons HCO_3^- + H^+ \rightleftharpoons CO_3^{2-} + 2H^+ \]

For pH values less than 8, typical of seafood, the concentration of carbonate ions may be neglected [136].

\[ CO_2 + H_2O \rightleftharpoons HCO_3^- \rightleftharpoons HCO_3^- + H^+ \]

According to Henry’s law, the concentration of CO₂ in the food is dependent on the water and fat content of the product, and on the partial pressure of CO₂ in the atmosphere [137]. The growth inhibition of microorganisms in MA is determined by the concentration of dissolved CO₂ in the product [138,139]. The preservation effect of MAP is due to the drop in surface pH in MA products because of the acidic effect of dissolved CO₂ but this could not entirely explain all of CO₂’s bacteriostatic effect [140]. The possibility of intracellular accumulation of CO₂ would upset the normal physiological equilibrium by slowing down enzymatic processes [140]. Thus, the effect of CO₂ on bacterial growth is complex and four mechanisms of CO₂ on micro-organisms has been identified [136,141-143]:

a. Alteration of cell membrane functions including effects on nutrient uptake and absorption
b. Direct inhibition of enzymes or decrease in the rate of enzyme reactions
c. Penetration of bacterial membranes, leading to intracellular pH changes
d. Direct changes in the physico-chemical properties of proteins

Probably a combination of all these activities account for the bacteriostatic effect. A certain amount (depending on the foodstuff) of CO₂ has to dissolve into the product to inhibit bacterial growth [144]. The ratio between the volume of gas and volume of food product (G/P ratio) should be usually 2:1 or 3:1 (greek food product). This high G/P ratio is also necessary to prevent package collapse because of the CO₂ solubility in wet foods. The CO₂ solubility could also alter the food-water holding capacity and thus increase drip [145].

Nitrogen (N₂) is an inert and tasteless gas, and is mostly used as a filler gas in MAP, because of its low solubility in water and fat [146,147]. N₂ is used to replace O₂ in packages to delay oxidative rancidity and to inhibit the growth of aerobic microorganisms, as an alternative to vacuum packaging. The use of oxygen in MAP is normally set as low as possible to inhibit the growth of aerobic spoilage bacteria. Its presence is reported to increase oxidative rancidity [121]. However, for some products oxygen could or should be used. High levels of oxygen are used in red meat and red fish meat, to maintain the red colour of the meat, to reduce and retard browning caused by formation of metmyoglobin [148]. O₂ in MA-packages of fresh fish will also inhibit reduction of TMAO to TMA [149].
Vacuum packaging and MAP, with high CO₂ levels (25-100%), extends the shelf-life of fish products from few days to a week or more compared with air storage, depending on species and temperature [89,135,141,150-157]. A shelf-life extension of 25-100% have been reported for fishery products [158-161]. Shelf life of some fishes under air, vacuum and MAP are given in Table 1.

Table 1: Shelf life of fishery products in air, vacuum and modified atmosphere packs.

<table>
<thead>
<tr>
<th>Type of Fish</th>
<th>Storage Temp (°C)</th>
<th>Atmosphere CO₂ : N₂ : O₂</th>
<th>Shelf Life (Days)</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Catfish fillets</td>
<td>4</td>
<td>Air</td>
<td>13</td>
<td>Reddy, Roman, Villanneva, Solomon, Kautter &amp; Rhodehamel [184]</td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>75 : 25 : 0</td>
<td>38-40</td>
<td></td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>Vacuum</td>
<td>20-24</td>
<td></td>
</tr>
<tr>
<td>Cod (Gadusmorhua) fillets</td>
<td>1</td>
<td>Air</td>
<td>9</td>
<td>Woyewoda, Bligh &amp; Shaw [185]</td>
</tr>
<tr>
<td></td>
<td>1</td>
<td>60 : 40 : 0</td>
<td>12</td>
<td></td>
</tr>
<tr>
<td>Cod (G. morhua) fillets</td>
<td>2</td>
<td>Air</td>
<td>11</td>
<td>Guldager, Bøknæs, Osterberg, Nielsen &amp; Dalgaard [186]</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>40 : 60 : 0</td>
<td>13</td>
<td></td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>40 : 40 : 20</td>
<td>20-24</td>
<td></td>
</tr>
<tr>
<td>Cod fillets</td>
<td>4</td>
<td>Air</td>
<td>20-24</td>
<td>Reddy, Solomon &amp; Rhodehamel [187]</td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>75 : 25 : 0</td>
<td>55-60</td>
<td></td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>Vacuum</td>
<td>24-27</td>
<td></td>
</tr>
<tr>
<td>Cod fillets</td>
<td>0</td>
<td>40 : 30 : 30</td>
<td>12.5</td>
<td>Cann et al. [158]</td>
</tr>
<tr>
<td></td>
<td>0</td>
<td>Vacuum</td>
<td>9</td>
<td></td>
</tr>
<tr>
<td>Cod (G. morhua) whole</td>
<td>2</td>
<td>100 : 0 : 0</td>
<td>10</td>
<td>Jensen, Petersen, Røgev &amp; Jepsen [188]</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>60 : 40 : 0</td>
<td>10</td>
<td></td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>40 : 60 : 0</td>
<td>9-10</td>
<td></td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>Vacuum</td>
<td>8-9</td>
<td></td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>Air</td>
<td>~7</td>
<td></td>
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<tr>
<td>Cod (G. morhua) whole/fillets</td>
<td>0</td>
<td>Air</td>
<td>12-13</td>
<td>Villemure, Simard &amp; Picard [189]</td>
</tr>
<tr>
<td></td>
<td>0</td>
<td>25 : 75 : 0</td>
<td>20</td>
<td></td>
</tr>
<tr>
<td>Cod (G. morhua) fillets</td>
<td>4</td>
<td>100 : 0 : 0</td>
<td>40-53</td>
<td>Post, Lee, Soberg et al. [190]</td>
</tr>
<tr>
<td>Cod, blue (Araperciscolias)</td>
<td>3</td>
<td>100 : 0 : 0</td>
<td>49</td>
<td>Penny, Belland Cummings [191]</td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>Vacuum</td>
<td>14</td>
<td></td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>Air</td>
<td>14</td>
<td></td>
</tr>
<tr>
<td>Haddock (Melanogrammus aeglefinus) whole</td>
<td>0</td>
<td>40 : 30 : 30</td>
<td>10</td>
<td>Dhananjaya &amp; Stroud [192]</td>
</tr>
<tr>
<td></td>
<td>0</td>
<td>Air</td>
<td>8</td>
<td></td>
</tr>
<tr>
<td>Haddock (M. aeglefinus) fillets</td>
<td>0</td>
<td>60 : 20 : 20</td>
<td>14</td>
<td>Dhananjaya &amp; Stroud [192]</td>
</tr>
<tr>
<td></td>
<td>0</td>
<td>Air</td>
<td>10</td>
<td></td>
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</tbody>
</table>
In spite of the various advantages, there are some limitations of vacuum and modified packaging method. The atmosphere within a vacuum and modified atmosphere pack may alter during storage due to reactions between components of the atmosphere and the product and/or due to transmission of gases in or out of the pack through the packaging film [162]. There is an increased concern about the growth/survival of micro-aerophilic psychrotrophic pathogens [163]. Modified atmosphere packaging usually extends the neutral taste period of fish rather than the initial fresh quality period [164]. Too high CO₂ concentrations in the atmosphere may have negative impact on drip loss, color, texture, and flavor in the product. These can be explained by the pH drop caused by CO₂ dissolving in the muscle tissue, resulting in a decrease in the water-holding capacity of the proteins, the denaturation of muscle and pigment protein, as well as the development of sour odor and flavor [79,165-167]. Due to the specific requirement of the consumers, diversity of product characteristics and basic fish packaging demands and applications, any packaging technologies that offer to deliver more product and quality control in an economic manner would be accepted universally. Active packaging is one such packaging approach currently exist which has great potential to substitute vacuum and modified atmosphere packaging technologies.

Active packaging: Over the past few years, there has been an increase in the demand for fresh, mildly preserved convenience foods that have better fresh-like qualities. In addition, changes in retail and distribution practices such as centralization of activities, new trends (e.g., internet shopping) and internationalization of markets, resulting in increased distribution distances and longer storage times of a set of different products with different temperature requirements, are putting huge demands on the food packaging industry. Traditional packaging concepts are limited in their ability to prolong the shelf-life of food products. This promoted the industry to develop new and improved methods for maintaining food quality and extending shelf life. Due to the complexities involved with fish products, many considerations are involved in choosing an acceptable packaging technology. One of the areas of research that has shown promise is that of active packaging (AP). Active packaging is an innovative concept that can be defined as a type of packaging that changes the condition of the packaging and maintains these conditions throughout the storage period to extend shelf-life or to improve safety or sensory properties while maintaining the quality of packaged food [168-170].

Active packaging (AP) performs some desired role other than providing an inert barrier between the product and external conditions [167,168,171-173], and combines advances in food technology, bio-technology, packaging and material science, in an effort to comply with consumer demands for ‘fresh like’ products [174]. Active packaging technique is either gas-flushing or more recently gas-scavenging or emitting systems added to emit (e.g., N₂, CO₂, ethanol) and/or to remove (e.g., O₂, CO₂, odour) gases during packaging or distribution. In case of a gas-scavenging or emitting system, reactive compounds are either contained in individual sachets or stickers associated to the packaging material or, more recently, directly incorporated into the packaging material. Major active packaging techniques are concerned with substances that absorb oxygen, ethylene, moisture, carbon dioxide, flavors/odours and those which release carbon dioxide, antimicrobial agents, antioxidants and flavors. Examples of currently known active packaging systems and their applications are given in Table 2. The most important active packaging concepts for fishery products include O₂ scavenging, CO₂ emitters, moisture regulators, antimicrobial packaging concepts, antioxidant release, release or absorption of flavors and odours.

Most widely used active packaging technology in the food industry is O₂ scavenger followed by antimicrobial packaging. For many foods, lipid oxidation is a major quality concern resulting in a variety of breakdown products which produce undesirable off-odours and flavors. Hence O₂ may cause off-flavors (e.g. rancidity as a result of lipid oxidation), colour changes (e.g. discoulouration of pigments such as carotenoids, oxidation), nutrient losses (e.g. oxidation of vitamin E, β-carotene, ascorbic acid) and accelerates microbial spoilage thereby causing significant reduction in the shelf life. Hence controlling O₂ assumes greater importance. Although this can be achieved by vacuum packaging and MAP, these technologies do not always remove O₂ completely. Moreover, the O₂ that permeates through the packaging film cannot be removed by these techniques. Use of an O₂ scavenger will help in solving these difficulties. O₂ scavengers were first commercialized by Japan’s Mitsubishi Gas Chemical Company (Ageless®) in 1970. O₂ scavengers are able to eliminate oxygen contained in the packaging headspace and in the product or permeating through

<table>
<thead>
<tr>
<th>Fish Species</th>
<th>O₂ Scavenger</th>
<th>CO₂ Scavenger</th>
<th>N₂ Scavenger</th>
<th>Ethanol Scavenger</th>
<th>O₂ Emitter</th>
<th>CO₂ Emitter</th>
<th>Ethanol Emitter</th>
</tr>
</thead>
<tbody>
<tr>
<td>Herring (Clupeaharengus) whole and fillets</td>
<td>00.0</td>
<td>60 : 40 : 0</td>
<td>14</td>
<td>Dhananjaya &amp; Stroud [192]</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>0</td>
<td>Air</td>
<td>12</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Salmon fillets</td>
<td>4</td>
<td>Air</td>
<td>24-27</td>
<td>Reddy, Solomon, Yep, Roman &amp; Rhodehamel [193]</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>75 : 25 : 0</td>
<td>55-62</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>Vacuum</td>
<td>34-38</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Snapper (Chrysophrysauratus) fillets</td>
<td>-1</td>
<td>Air</td>
<td>9</td>
<td>Scott, Fletcher &amp; Hogg [194]</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>-1</td>
<td>40 : 60 : 0</td>
<td>9</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>-1</td>
<td>100 : 0 : 0</td>
<td>18</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

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Table 2. The most important active packaging concepts for fishery products include O₂ scavenging, CO₂ emitters, moisture regulators, antimicrobial packaging concepts, antioxidant release, release or absorption of flavors and odours.
the packaging material during storage. O2 scavengers are widely used to slow down or to prevent deterioration due to product components oxidation and/or growth of microorganisms or survival of insects [175]. O2 scavengers are efficient in preventing discoloration of cured meats and tea, rancidity problems in high fat foods including fishes, mould spoilage of intermediate and high moisture bakery products or oxidative flavor changes in coffee [176-179]. O2 scavenging concepts are mainly based on, iron powder oxidation, ascorbic acid oxidation, photosensitive dye oxidation, enzymatic oxidation (e.g. glucose oxidase and alcohol oxidase), unsaturated fatty acids (e.g. oleic or linolenic acid), rice extract or immobilized yeast on a solid substrate [180]. The main cause of spoilage of many foods is microbial growth on the product surface. Antimicrobial packaging, which are grouped among active packaging, contains small amounts of natural or synthetic antimicrobial agent. The strategy of antimicrobial packaging film depends on release of antimicrobial agent incorporated into a packaging material on to food surface. Thus, antimicrobial packaging film may delay or even prevent the growth of microorganisms on the product surface leading to extension of the shelf life with improved safety. As in MAP foods, the function of carbon dioxide within a packaging environment is to suppress microbial growth to extend the shelf life. CO2 has a prevailing inhibitory effect on bacterial growth. It is particularly effective against gram-negative, aerobic spoilage bacteria such as pseudomonas species that causes off-colour and flavors in fish [130]. The overall effect of CO2 is to increase both the lag phase and the generation time of spoilage microorganisms. Over the years this has been achieved by modified atmosphere packaging, in which a package is flushed with a mixture of gases including carbon dioxide at sufficient levels. However the concentration of CO2 in the MAP foods will vary during storage period which lead to the development of CO2 emitters which can emit continuously throughout the storage period. Various chemical combinations like sodium bicarbonate, citric acid, sodium dihydrogen phosphate, ferrous carbonate etc have been used to develop CO2 emitters [181]. Considerable shelf life advantage was obtained for Seer fish packed under CO2 emitters [181-183]. Shelf life of fishery products in active packaging systems are given in Table 3. Other active packaging systems that are expected to find increased attention in the future include colour containing films, light absorbing or regulating system, susceptors for microwave heating, gas permeable/breathable films, anti-fogging films and insect repellant packages.

Table 2: Examples of some currently known active packaging systems and their applications in food systems.

<table>
<thead>
<tr>
<th>Type of Active Packaging System</th>
<th>Substances Used</th>
<th>Applications</th>
</tr>
</thead>
<tbody>
<tr>
<td>O2 absorbing</td>
<td>Chemical systems (powdered iron oxide, catechol, ferrous carbonate, iron-sulfur, sulfite salt-copper sulfate, photosensitive dye oxidation, ascorbic acid oxidation, catalytic conversion of oxygen by platinum catalyst)Enzymatic systems (glucose oxidase-glucose, alcohol oxidase-ethanol vapour)</td>
<td>Fresh and dry fish, sausages, smoked and cured meats, roasted nuts, coffee, cereals, chocolate, cheese, bread, cakes, pastries, fruit juice, ready to drink tea, beer, wine,</td>
</tr>
<tr>
<td>CO2 absorbing</td>
<td>calcium hydroxide, sodium hydroxide or potassium hydroxide</td>
<td>Roasted coffee, fruit, cheese, poultry products</td>
</tr>
<tr>
<td>CO2 emitting</td>
<td>Ascorbic acid, ferrous carbonate, metal halide</td>
<td>Fresh fish and meat, nuts, potato chips</td>
</tr>
<tr>
<td>Moisture absorbing</td>
<td>Silica gel, propylene glycol, polyvinyl alcohol, diatomaceous earth</td>
<td>Fish and meat, all dry products, cheese, bread, biscuits</td>
</tr>
<tr>
<td>Ethylene absorbing</td>
<td>Activated charcoal, silica gel-potassium permanganate, kieselsiure, bentonite, fuller’s earth, silicon dioxide powder, powdered oya stone, zeolite, ozone</td>
<td>Fruits and vegetables</td>
</tr>
<tr>
<td>Ethanol emitting</td>
<td>Encapsulated ethanol</td>
<td>Fresh and Semidry Fish, cheese, high moisture bakery products</td>
</tr>
<tr>
<td>Antimicrobial releasing</td>
<td>Sorbates, benzoates, propionates, ethanol, ozone, peroxide, sulfur dioxide, antibiotics, silver-zeolite, quaternary ammonium salts</td>
<td>Fish, meat, cheese, fruits, bread, cakes</td>
</tr>
<tr>
<td>Antioxidant releasing</td>
<td>BHA, BHT, TBHQ, ascorbic acid, tocopherol</td>
<td>Fish, cereals,</td>
</tr>
<tr>
<td>Flavor absorbing</td>
<td>Baking soda, active charcoal</td>
<td>Fish, fruit juices</td>
</tr>
<tr>
<td>------------------</td>
<td>-----------------------------</td>
<td>-------------------</td>
</tr>
<tr>
<td>Flavor releasing</td>
<td>Many food flavors</td>
<td>Fish, ground coffee, ice cream</td>
</tr>
<tr>
<td>Colour containing</td>
<td>Various food colours</td>
<td>Surimi</td>
</tr>
<tr>
<td>Anti-fogging and anti-sticking</td>
<td>Biaxially oriented vinylon, compression rolled oriented HDPE</td>
<td>Fresh fruits and vegetable</td>
</tr>
<tr>
<td>Light absorbing / regulating</td>
<td>UV blocking agents, hydroxyl benzophenone</td>
<td>Milk, pizza</td>
</tr>
<tr>
<td>Microwave susceptors</td>
<td>Metalized thermoplastics</td>
<td>Ready to eat meals</td>
</tr>
<tr>
<td>Insect repellant</td>
<td>Low toxicity fumigants (pyrethrins, permethrin)</td>
<td>Dry fish, cereals</td>
</tr>
</tbody>
</table>

Table 3: Shelf life of fishery products stored under different packaging conditions.

<table>
<thead>
<tr>
<th>Type of Fish</th>
<th>Storage Temp (°C)</th>
<th>Packaging Atmosphere</th>
<th>Shelf Life (Days)</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Catfish steaks</td>
<td>0-2</td>
<td>Air</td>
<td>10</td>
<td>Mohan et al. [90]</td>
</tr>
<tr>
<td></td>
<td></td>
<td>O₂ Scavenger</td>
<td>20</td>
<td>Mohan, Ravishankar, Srinivasa Gopal and Ashok Kumar [182]</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Mohan, Ravishankar et al. [179]</td>
</tr>
<tr>
<td>Seer fish steaks</td>
<td>0-2</td>
<td>Air</td>
<td>12</td>
<td>Mohan, Ravishankar et al. [157]</td>
</tr>
<tr>
<td></td>
<td></td>
<td>O₂ Scavenger</td>
<td>20</td>
<td>Mohan, Ravishankar et al. [183]</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Control O₂ Scavenger</td>
<td>12</td>
<td>Mohan, Ravishankar et al. [183]</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Sodium acetate treated O₂ Scavenger</td>
<td>25</td>
<td>Mohan, Ravishankar et al. [183]</td>
</tr>
<tr>
<td>Indian Oil Sardine</td>
<td>0-2</td>
<td>Air</td>
<td>5</td>
<td>Mohan, Ravishankar et al. [183]</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Edible chitosan coating</td>
<td>~8-10</td>
<td>Mohan, Ravishankar et al. [183]</td>
</tr>
</tbody>
</table>

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