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Final Project 2023

APPLICATION OF QUALITY INDEX METHOD (QIM) SCHEME FOR SUB CHILLED FARMED ATLANTIC SALMON (*Salmo salar*)

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This paper should be cited as: Nirbadha, S. 2024. *Application of Quality Index Method (QIM) scheme for sub-chilled farmed Atlantic salmon (Salmo salar).* GRÓ Fisheries Training Programme under the auspices of UNESCO, Iceland. Final project.
https://www.grocentre.is/static/gro/publication/1961/document/Nirbadha23prf.pdf

ABSTRACT

The quality of fresh fish is a concern for both the fishing industry and consumers in Sri Lanka due to insufficient cooling practices and limited quality inspection. Sensory evaluation plays an important role in assessing fish freshness and supporting quality control. This study evaluated quality changes and shelf life of gutted farmed Atlantic salmon (*Salmo salar*) stored under sub-chilled (SUBC) and ice-chilled (ICEC) conditions and assessed whether modifications to the Quality Index Method (QIM) scheme are required for sub-chilled salmon. A total of 54 salmon (average weight 4.42 ± 0.73 kg) were divided into two groups (n = 27 each). The SUBC group was stored in ice at -2°C to 0°C until day 16 and then at 0°C to 4°C until day 23, while the ICEC group was stored at 0°C to 4°C throughout the 23-day period. Three fish from each group were sampled on nine storage days (1, 4, 8, 11, 14, 16, 18, 21, and 23). Sensory quality of whole fish was assessed using QIM, cooked samples were evaluated using Generic Descriptive Analysis (GDA), and microbial quality was determined by Total Viable Counts (TVC). Temperature loggers monitored storage conditions throughout the experiment. QIM results showed that quality index scores increased linearly with storage time in both groups. Scores for skin odour, skin texture, eye form, gill mucus, blood in the abdomen, and abdomen odour were similar between treatments. However, SUBC salmon consistently received higher scores for eye pupil and eye form attributes, while ICEC salmon showed higher scores for skin mucus and gill colour after day 16. GDA indicated a decline in freshness characteristics over time, but no spoilage attributes were detected. The only significant sensory difference was observed on day 23, when SUBC salmon displayed a fresher flavour than ICEC salmon. TVC remained within acceptable limits in SUBC salmon, whereas ICEC salmon reached unacceptable microbial levels on days 21 and 23. The results indicate that gutted farmed Atlantic salmon stored under sub-chilled conditions have a shelf life exceeding 23 days in ice storage, while conventionally iced salmon exhibit a shorter shelf life. No adjustment of the QIM scheme was considered necessary for sub-chilled salmon.

Key words: Atlantic salmon (*Salmo salar*), subchilling, ice-chilling, Quality Index Method (QIM), shelf life, sensory evaluation, Sri Lanka.

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1 INTRODUCTION

Sri Lanka is a small tropical island in the Indian Ocean, situated off the southern tip of India, with an exclusive economic zone covering approximately 517,000 km^2 (Jianadasa, *et al*., 2014). In 2021, the annual capture fisheries production was 331,675 metric tons, and the total annual fisheries production, including aquaculture, was 435,910 metric tons (State Ministry, 2021). These statistics indicated a 1.5% increase in seafood production compared to the previous year. The total contribution of the fisheries subsector to the gross domestic product (GDP) was 1.1% in 2021 (DFAR, 2022). Owing to its significant contribution to the national economy, efforts are being made to further increase the fisheries sector's contribution to GDP (World Bank, 2021). As a result of some policy changes, the export earnings for fish and fishery products reached approximately 26,749 metric tons in 2021, generating a record-high export income of around $318 million compared to previous years (State Ministry, 2021). Additionally, the fisheries sector provides nearly 586,000 employment opportunities across the country (DFAR, 2022).

Marine capture fisheries in Sri Lanka are broadly categorised into coastal and offshore fisheries, with the coastal fisheries further divided into pelagic and demersal subsectors. Seasonal demersal fisheries are prevalent in most coastal areas of Sri Lanka. Over 15 fishery resource surveys have been conducted in Sri Lankan waters since 1920, predominantly focusing on demersal resources (Samaranayake, 2003). Some of these surveys estimated the potential yield of the demersal fishery in Sri Lanka. For instance, a survey conducted by the Dr. Fridtjof Nansen research vessel (RV) between 1978 and 1980 estimated demersal fishery resources at 80,000 t (Samaranayake, 2003). After 40 years, in 2018, another survey was conducted by RV Dr. Fridtjof Nansen in Sri Lankan waters. This survey recorded four main families of demersal species (Carangidae, Lethrinidae, Lutjanidae, and Serranidae) with great potential for demersal fisheries. The estimated total biomass for each family was 2, 695 t, 1, 876 t, 2, 720 t, and 1, 102 t for Carangidae, Lethrinidae, Lutjanidae, and Serranidae, respectively (Krakstad *et al*., 2018). Additionally, the survey revealed a high potential for demersal fishery in Sri Lanka, particularly for species such as groupers, snappers, and sweetlips, owing to the lower fishing pressure (Gunasekara *et al*., 2019). These demersal fish species, including groupers and snappers, are categorised as high-demand species among both local and foreign consumers, especially in tourism areas of Sri Lanka (Samaranayake, 2003).

When considering the demersal fishery of Sri Lanka, most fishermen engage in fishing activities using outer-fitted engine fibreglass-reinforced boats (OFRP), employing main fishing gear types such as gill nets and purse seines. Typically, fishermen return to shore after 6–8 hours of fishing, and the catch is packed in polystyrene boxes with ice. However, if fish are caught during the daytime, they are exposed to the sun and wind, leading to increased core temperatures and susceptibility to spoilage. Demersal fish species in Sri Lanka have high fat and water content (normally 1-24% fat and 60-81% water), making them susceptible to spoilage if not cooled promptly after capture (Mallikage, 2001). Proper handling and storage of fish at sea are essential to ensure that the catch remains as fresh as possible until it is landed. An effective on-board handling system is required to cool fish to a storage temperature just above the freezing point.

In Sri Lanka, the coastal fishery fleet mainly uses polystyrene boxes with crushed ice to cool fish on board. However, this practice is not universal among boats engaged in demersal fisheries. Consequently, the delayed cooling of fresh fish can significantly impact its quality. To enhance fish freshness, the Sri Lankan government has regulated two main storage

conditions: chilled and frozen. According to the amendment published in 2020 under the Food Act No. 26 of 1980, chilled fresh fish should be cooled to a temperature approaching the melting ice range of -1°C to 4°C. Chilled fish should not be frozen, and the water should remain in the liquid stage, while the chilled fish should maintain visual quality. Frozen fresh fish undergo freezing until they reach a core temperature of -18°C (Gazette, 1980). However, these regulated conditions are often not followed in the local retail market throughout the entire value chain until reaching the consumer level, except by a few reputable seafood processing companies in Sri Lanka. The quality of fresh fish is an important concern for both the fish industry and consumers.

Atlantic salmon (*Salmo salar*) is in significant demand among consumers worldwide, contributing to a global salmon consumption three times higher than that of the 1980s. It is considered a luxury food and ranks among the most popular fish species in the US, Europe, and Japan. Salmon aquaculture is the fastest-growing food production system globally, capturing 70% of nearly 2.5 million metric tons of the market. According to the FAO's State of World Fisheries and Aquaculture report, Atlantic salmon and 21 other dominant species, such as milkfish, collectively account for 75.6% of all finfish species in mariculture and coastal aquaculture. With a production of 2.7 million tons in 2020, Atlantic salmon alone represented a significant 32.6% share of marine and coastal aquaculture of all finfish species.

Freshness is the most important attribute in evaluating fish quality (Osibona and Ezekiel, 2014). Various sensory evaluation methods exist to estimate the freshness and shelf life of whole fish and fillets. Among these, the European Union (EU) scheme, which includes the Quality Index Method (QIM) and Torry schemes (Martinsdottir, *et al.*, 2001), is widely recommended and documented. The Torry scheme, prevalent in the fish industries of several countries, assesses the overall freshness of cooked fillets based on odour and flavour characteristics. Conversely, the QIM method is esteemed for its cost-effectiveness, as it requires no specialised equipment and relies solely on trained sensory assessors to perform the evaluations. Moreover, the QIM method is renowned for its swiftness, reliability, and noninvasive nature, making it an ideal tool for fish quality control management. However, it must be adjusted according to the sensory characteristics of each species.

Recent studies have focused on developing Quality Index Method (QIM) schemes tailored to individual fish species. These endeavours necessitate specific knowledge about fish species, trained sensory assessors, and standardised storage experiments. In Sri Lanka, several QIM scheme sensory evaluation studies have been conducted for pelagic fish species, including spotted sardinella (*Amblygaster sirm*) (Perera *et al*., 2020), frigate tuna (*Axis thazard*) (Ariyawansa *et al*., 2003), and various tuna species (frigate and yellowfin tuna) (Edirisinghe *et al*., 2019). However, there is a notable absence of published QIM scheme sensory evaluation literature for common demersal fish species in Sri Lanka.

Given the high demand for demersal fish species in Sri Lanka, ensuring the freshness quality of fish is paramount for both industrial and consumer ends in the country. Unfortunately, limited attention has been given to developing knowledge for identifying the freshness quality of fish using the QIM scheme sensory evaluation. Therefore, it is imperative to develop QIM schemes for sensory evaluation tailored to the demersal fish species in Sri Lanka. Sensory evaluation is the most practical, quick, reliable, and cost-effective method for developing countries such as Sri Lanka.

The primary objective of this project is to gain knowledge and experience in the sensory evaluation of fish, particularly focusing on freshness assessment and shelf-life studies of fish stored under sub-chilled and ice-chilled conditions. This will involve the assessment of a Quality Index Method (QIM) scheme to evaluate freshness and conduct shelf-life studies of fish, particularly targeting commercially important diadromous fish species in Iceland.

The acquired knowledge and experience will be utilised to develop QIM schemes tailored for commercially significant demersal fish species in Sri Lanka. Furthermore, the expertise gained will be leveraged to train a team of sensory panellists, such as staff members of the Department of Fisheries and Aquatic Resources (DFAR) and the National Aquatic Resources Research and Development Agency (NARA). This aspect of the project holds significant value for developing countries such as Sri Lanka, providing a method that is quick, applicable, reliable, and cost-effective.

1.1 Project summary

The overarching objective of this project was to explore the application of sensory evaluation in the Sri Lankan demersal fishery industry. This study involved the application of a sensory evaluation method to assess fish freshness, specifically utilising the developed Quality Index Method (QIM) in a comprehensive shelf-life study. This study evaluated the need to adapt the QIM scheme developed for iced whole farmed Atlantic salmon (*Salmo salar*) to sub-chilled conditions, employing trained panellists at Matis for evaluation in the shelf-life study. The knowledge and technology garnered from this endeavour will be transferred to demersal fishery species in Sri Lanka, facilitating the development of QIM schemes tailored for each species under ice storage conditions.

1.2 Problem statement and background

In 1920, Le Danois described the super-chilling process for the first time, although the term "super-chilling" or "deep-chilling" was not used (Zhou *et al*., 2010). The novel technology was introduced based on the concept of sub-chilling, aiming to address the issue of temperature fluctuation and lack of control in the processing of fresh salmon within the value-added supply chain. The implementation of this technology was expected to result in improved quality of fresh products and longer shelf-life, increased security in the value-added cold chain, and reduction in production and logistics costs. However, despite advancements in technology, there is a lack of simplified methods for producers and consumers to assess the quality of fresh products, especially when compared to chemical or sophisticated instrumental (liquid/gas chromatographic) food analysis methods. Therefore, sensory evaluation is an ideal tool for identifying the quality of fresh products, given its applicability, speed, reliability, and cost-effectiveness, particularly in developing countries such as Sri Lanka.

Furthermore, producers in Iceland, such as the salmon producer Arnarlax, have requested that Matis revise or develop the existing Quality Index Method (QIM) for farmed Atlantic salmon in ice for sub-chilled storage conditions. Arnarlax agreed to provide salmon samples free of charge to facilitate the adjustment of the QIM for cultured Atlantic salmon in sub-chilled storage. Arnarlax's expectation is the creation of a more accurate and reliable QIM for cultured Atlantic salmon under sub-chilled conditions, along with well-explained publications on sensory evaluation to assess the quality of the flesh and shelf life of the fish. Therefore, it is essential to utilise trained sensory panellists for the development of QIM for farmed Atlantic salmon under sub-chilled conditions.

Expanding the quality of flesh and shelf-life of farmed Atlantic salmon using sub-chilled conditions will be advantageous for stakeholders in the country. Currently, they must spend a significant amount of money on air freight to distribute their products worldwide to maintain high-quality standards. The aim of the project is to evaluate whether adjustments of the QIM scheme are needed for subchilled salmon and conduct a shelf-life study comparing the quality attributes of salmon stored in ice and sub-chilled. The objective is to ensure that the freshness of salmon could be accurately evaluated at any point during transport to the market. The aim of this project is to evaluate the shelf life of salmon using sub-chilled storage. Such information can be used by stakeholders to support the transition from air freight to shipping freight for transportation worldwide. The ultimate results of the project will be advantageous to both producers and consumers, particularly Arnarlax, as they seek accurate measures of the shelf life of their sub-chilled salmon production. The ultimate results of the project will be advantageous to both producers and consumers, as it will reduce transportation costs by switching from air freight to shipping freight.

1.3 Project objectives

1. Assess the need to adjust the Quality Index Method (QIM) scheme for sub-chilled farmed Atlantic Salmon (*Salmo salar*).
2. Conduct a shelf-life study of sub-chilled and ice-chilled whole fresh farmed Atlantic Salmon (*Salmo salar)* stored in ice using the QIM scheme, Generic Descriptive Analysis (GDA), and microbiological methods.

2 LITERATURE REVIEW

2.1 Farmed Atlantic salmon - *Salmo salar* (Linnaeus, 1758) and aquaculture

Salmo salar, known as Atlantic salmon, belongs to the Salmonidae family and inhabits marine, freshwater, and brackish water environments. These salmon are categorised as demersal or benthopelagic species because they occupy seabeds and river floors. Due to their spawning behaviour, they are categorised as anadromous fish species. Atlantic salmon are distributed naturally in the North Atlantic Ocean, temperate and arctic zones in the Northern Hemisphere, and the Western Atlantic. In addition, Atlantic drainages from northern Quebec, Canada, to Connecticut and New York, USA, geographically between 72°N - 37°N to 77°W - 61°E. (Froese and Pauly, 2024). Biologically, amphihaline species spend most of their lives in freshwater. They remain in freshwater for their first–6 years but then migrate to coastal marine waters or even to open oceans, where they remain for 1–4 years before returning to freshwater for spawning. After 1-4 years at sea, they migrate back to the upper reaches of their natal rivers to spawn.

Atlantic salmon farming began in the 19th century in the United Kingdom in freshwater as a means of stocking waters with parr to enhance wild returns for anglers. Marine cage culture was first used in the 1960s in Norway for fattening salmon to a marketable size. (FAO Factsheet, 2009) Fishing pressure on wild stocks has decreased due to intensive farming but other problems have increased. According to the FAO (2022), the dominant species in marine culture is salmon, which contributes to the global production of aquatic animals. In 2020, the worldwide production of salmon was 2719.6 thousand tonnes of live weight, representing 32.6 percent of the total. Furthermore, the report mentioned that Atlantic salmon, with a production of 2.7 million tonnes in 2020, accounted for a high 32.6 percent of marine and coastal aquaculture among all finfish species.

Aquatic foods play a crucial role in maintaining a healthy and balanced diet. Even small quantities of aquatic foods can significantly improve nutrition by providing essential nutrients that may be scarce in plant-based diets. These foods offer high-quality proteins, essential amino acids, vitamins (particularly A, B, and D), phosphorus, and minerals, such as iron, calcium, zinc, iodine, magnesium, potassium, and selenium. They also serve as a primary dietary source of heart-healthy omega-3 fatty acids (FAO, 2022). The nutrient content of aquatic foods varies depending on the species. One significant distinction lies in fat content: species such as sardines, salmon, and tuna are considered fatty, whereas cod and catfish are lean. The two omega-3 fatty acids found in aquatic species are eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA). As the human body does not produce omega-3 fatty acids, they must be obtained through food. Although omega-3 fatty acids are present in all types of aquatic food, they are especially abundant in fatty fish. Regular consumption of aquatic food contributes to maintaining a healthy heart by reducing blood pressure and lowering the risk of stroke, depression, Alzheimer's disease, and other chronic conditions.

According to FAO (2022), nearly 90 percent of the quantity (live weight equivalent) of traded aquatic products consists of preserved products, with the majority being frozen. However, owing to the demand for fresh aquatic products and advancements in packaging and logistical technologies, the proportion of fresh products in trade volumes has increased over time. In 1976, fresh products accounted for 5.8 percent of the quantity (live weight) of traded aquatic products, compared to 11.1 percent in 2020. The development of airfreight has facilitated the

export of high-end fresh aquatic products such as farmed salmon and captured demersal finfish species.

2.2 Fish storage techniques in ice

Freshly caught fish are prone to spoilage and require processing and preservation to extend their shelf life. Preservation aims to maintain the quality of fish and fish products, particularly by preventing microbial spoilage. According to Gökoğlu and Yerlikaya (2015), preservation involves storing excess fish, caught abundantly or produced in surplus, to be consumed fresh during times of scarcity or when transported over long distances. Preservation methods vary in terms of their effectiveness and the duration of shelf-life extension. The choice of method depends on factors such as the product type, properties, available energy sources, storage facilities, and cost considerations (Gökoğlu and Yerlikaya, 2015). In some cases, combining preservation methods may be necessary to achieve optimal results. Typically, fish preservation begins on fishing vessels, where immediate cooling is essential to counteract accelerated spoilage caused by high ambient temperatures. Chilling, refrigeration, and freezing are common preservation methods used on board vessels and inshore facilities. Fish are transported to land under cold conditions and stored in cold storage facilities until they are further processed or marketed (Gökoğlu and Yerlikaya, 2015).

2.2.1 Ice chilling

Chilling is employed to lower the temperature of fish to approximately 0ºC. Although chilling slows the spoilage process, it does not completely halt it. The effectiveness of chilling depends on various factors, including the initial microbial load, chemical composition of the fish, temperature, relative humidity, and air velocity (Gökoğlu & Yerlikaya, 2015). Chilling is particularly effective at inhibiting the growth of mesophilic and thermophilic microorganisms. According to Gökoğlu and Yerlikaya (2015), one of the most common and efficient chilling methods involves the use of ice. In this method, the fish is fully surrounded by ice, leveraging its excellent cooling capacity. As ice melts, it absorbs heat from the fish, effectively cooling it. Additionally, chilled or refrigerated seawater (RSW) is used for fish chilling, a practice commonly observed in onboard applications (Gökoğlu and Yerlikaya, 2015).

2.2.2 Sub-chilling

In 1920, Le Danois described the super-chilling process for the first time, although the term "super-chilling" or "deep-chilling" was not used (Zhou *et al*., 2010). Generally, super chilling implies temperatures at the borderline between conventional chilling and freezing. The super-chilling process has been defined differently by several researchers. Ando *et al*. (2004) defined it as the temperature zone below 0ºC, where ice crystals are not formed. The concept of super chilling technology was described by Beaufort *et al*. (2009), who reported that food is stored just below its initial freezing temperature, or more precisely, the temperature of the food product is lowered to 1–2 °C below its initial freezing point (Duun and Rustad, 2007). Magnussen et al. (2008) described the terms "sub-chilling", "partial-freezing", or "shell-freezing" as a process in which a minor part of the product's water content (5%–30%) is frozen. After initial surface freezing (1–3 mm layer of ice), the formed ice absorbs heat from the food interior, eventually leading to an equilibration of temperature between the interior and exterior during food storage and distribution (Magnussen *et al*., 2008).

According to Kaale *et al*. (2011), the super-chilling method comprises two primary phases: the super-chilling process and super-chilled storage. The superchilling process involves partial freezing, resulting in ice formation along the edges of the product. Subsequently, during super-

chilled storage, the product is kept at temperatures lower than the conventional chilling range of 4–10ºC. The super-chilling process can be delineated into two stages. First, the precooling stage entails cooling the product to its initial freezing point. Second, the latent heat of crystallisation is removed.

The primary reason for implementing super-chilling technology is its ability to combine the favourable effects of low temperature with the conversion of some amount of free water into ice (Kaale *et al*., 2014), thereby making it less available for deteriorative processes (Banerjee and Maheswarappa, 2017). For shorter storage or transport periods, this process provides a refrigeration reservoir, eliminating the need for external ice around the product (Wang *et al*. 2008). This approach reduces the amount of ice used during the supply chain, minimising transport weight and costs, and has a positive impact on the environment (Bahuaud *et al*., 2008). It has been used for the preservation of salmon fillets and seabass throughout the distribution chain by utilising chambers with controlled temperatures of -1 to -3ºC without ice (Chang *et al*., 1998; Huss, 1998). However, for long-term storage, refrigeration under super-chilling conditions (1–1.5 °C below the product's freezing point) is necessary. The shelf life of superchilled food is shorter than that of frozen food but longer than that of chilled food (Banerjee and Maheswarappa, 2017). Compared to traditional chilling, it improves the shelf life of foods by 1.5 to 4 times (Kaale *et al*., (2011). Gallart-Jornet *et al*. (2007) reported superchilling as a promising method for minimising biochemical degradation, protein denaturation, and structural damage in Atlantic salmon compared to frozen storage. However, the main concern of this technology is the formation and growth of ice crystals (Einarsson, 1988, Duun and Rustad, 2008). At super-chilling temperatures, microbial activity is reduced, and most bacteria cannot grow (Kaale, *et al*., (2011), but autolytic, enzymatic, or other chemical reactions may occur at a higher rate (Einarsson, 1988) or even accelerate, as reported by Magnussen, *et al*., (2008).

For meat, fish, and seafood products, this technology will be a boon in extending their shelf life. In addition to the cost of manpower and unit operations for freezing-thawing, the weight loss of the products during these processes results in significant economic loss. As a commercial practice, super-chilling can reduce the need for freezing/thawing of production buffers, minimise labour and energy costs, and improve product yield (Zhou *et al*., 2010). It may also reduce the processing time, thereby increasing the capacity of the chilling equipment. Superficial ice formation in superchilled products protects them from temperature abuse.

Banerjee and Maheswarappa (2017) also mentioned that, in addition to improving shelf life, companies claim that the super-chilling process can reduce energy use and waste. The energy required to produce and distribute chilled and superchilled salmon was calculated. Although more energy was required for super-chilling fish during manufacture, more fish could be packed into each vehicle, and as no extra ice was required during transportation, the number of journeys required was reduced. The extra energy used during the super-chilling process was equated to fuel savings. The food industry is under constant pressure to produce fresh, additive-free fish and meat products with extended shelf lives, and superchilling offers a cost-effective and simple method for achieving this target. Research and trials so far indicate that some products respond more favourably to super-chilling than others, such as fish, seafood, and meat, in which the extensive use of this technology has shown promising results.

Some scientific investigations have revealed several advantages of using the super-chilling process industrially. Extending the shelf life of fresh food products is essential in the industry. Delivering food via air transport while keeping it fresh incurs costs both in terms of

environmental and monetary costs. However, with super-chilling, products can be transported by ships and arrive fresh. This is in contrast to chilled products, which have such a short shelf life that they must be transported by air (Banerjee and Maheswarappa, 2017).

Freezing is another widely-used conservation method industrially, but it has downsides. When thawed, the product is no longer fresh, and there is an additional economic cost of freezing/thawing due to the weight loss of the product (Banerjee and Maheswarappa, 2017). However, according to Magnussen *et al*. (2008), the industrial use of super-chilling can decrease the use of freezing/thawing for production buffers. This reduction can lead to savings in energy costs, labour, and product weight losses, thereby improving the yield (Zhou, *et al*., 2010). Additionally, it can decrease the processing time, which increases the capacity of the chilling equipment. Some superficial ice formation in super-chilled foodstuffs helps protect them from temperature abuse (Banerjee and Maheswarappa, 2017).

Although more energy is needed to manufacture super-chilled products, more fish can be packed into each transport vehicle as no extra ice is needed during transportation (Banerjee and Maheswarappa, 2017). The transport of food products is more effective when there is no external ice, as the products can be packed tighter, weigh less, and there is no latent spill water during transport. In traditional chilling, ice adds an extra approximate 20-30% of the weight of each box of fish (Magnussen *et al*., 2008), which is avoided with the use of super-chilling. Here, the refrigeration capacity is stored within the product owing to the thin lining of ice along the product's edges, and the need for external ice is thus eliminated. Using super-chilling could theoretically reduce the number of trucks exporting salmon from Norway to Europe by 20-30% (Magnussen *et al*., 2008). However, there are requirements regarding whether the product is intended to be further stored as super-chilled or to be thawed and stored as chilled. Furthermore, a break in the cold chain leading to an increased product temperature would also mean that it would take longer for the product to return to the desired temperature (Duun, 2008). No ice should form on the surface of the products during traditional chilling processes. Therefore, the driving force for heat removal is very low, particularly for thick products, where internal heat transfer is slow owing to low thermal conductivity and a long thermal conduction path. Consequently, traditional industrial chilling is time-consuming. For comparison, a common size salmon (4–5 kg) can be chilled from 15 to 2°C in approximately 2 h at -1°C (Magnussen *et al*., 2008). The super-chilling process (partial freezing) can be applied both before traditional chilled distribution or before super-chilled storage, where the refrigeration capacity is stored in products. It can also be applied to fishing in offshore waters, where there is insufficient ice to maintain the quality of the catch.

2.3 Fish freshness and shelf life

Fish freshness is a crucial factor in ensuring food safety. Maintaining high-quality fish throughout the complex chain from catch to consumption is essential. Botta (1995) cited 15 different definitions of quality, ranging from general statements to consumer perspectives, highlighting the complexity of defining quality. Nielsen and Hyldig (2004) emphasised that quality cannot be simply defined and varies depending on the context and species, influenced by biological and technical parameters. We agree with this perspective and believe that chemical parameters should also be considered.

Seafood is highly perishable (Defoe, 2015). Martinsdóttir *et al*. (2002[a]) noted that the deterioration process begins as soon as seafood is caught, affecting its quality as a food product. Changes in the composition and structure of seafood due to biochemical, physical, enzymatic,

and bacterial processes negatively impact its sensory quality. Bremner (2000) described quality as a multidimensional concept influenced by various parameters affecting a product's quality. However, producing high-quality food requires detailed information on the quality of raw materials and products.

Most of the super-chilled studies highlighted farmed Atlantic salmon as a rich source of essential polyunsaturated fatty acids, potentially enhancing the nutritional level of the growing population. However, Atlantic salmon tends to have a shorter shelf life than most other food products because of its high polyunsaturated fatty acid content and susceptibility to microbial spoilage. Earlier studies have indicated that the use of sub-chilling can extend the shelf life of Atlantic salmon Gökoğlu and Yerlikaya (2015).

Quality changes during the storage of salmon occur due to various biological, chemical, and physical factors affecting the body of the salmon, both externally and internally. These factors include drip loss, water content, cell tissue fluid, pH, temperature, colour, microbial growth, primary and secondary oxidation product content, and protein solubility. Biologically affecting factors associated with cultured salmon include the size of the fish, sex, maturity stage, stress before slaughter, parasitic attachment, fish tissue anatomy and structure, rigor stage, muscle thickness, and lag phase of spoilage microbes. Additionally, chemical factors such as the fish feeding regime during aquaculture, nutritional composition, protein content, fat content, water content, oxidation states, protein denaturation, and pH in the muscle tissue play significant roles. Physical factors, such as temperature, cell tissue fluid, and drip loss, also have a notable impact. These factors directly or indirectly influence the quality of the fish and the shelf life of the salmon. (Doyle, (1989); Sveinsdottir, *et al*., 2002; Martinsdottir, *et al*., 2003; Bahuaud, *et al*., (2008); Duun and Rustad, (2008); Sebra and Pedrosa, (2010); Kaale, (2011 and 2013); Gökoğlu, and Yerlikaya, (2015); Kaale and Eikevik, (2016); Ochiai and Ozawa, (2020)).

Superchilling prolongs the shelf life of food compared to conventional chilling (Duun, 2008). Furthermore, the shelf life of super-chilled products is much lower than that of frozen products, as a larger proportion of the bulk water remains unfrozen compared to frozen products. However, there is less structural damage in super-chilled foods because of a lower degree of freeze denaturation and drip loss. Kaale and Eikevik (2016) found that super-chilled foods have many advantages over traditional chilling or freezing. At traditional chilling temperatures, many spoilage microorganisms are highly active, and pathogenic bacteria can grow and produce toxins. Therefore, it is necessary to lower the temperature as much as possible without damaging the product. This is done in super-chilling, where a mix of partial freezing and storage at low temperatures extends the shelf life of the product by drastically reducing microbial activity (Duun and Rustad, 2008).

The initial freezing point of most foods typically falls within the range of -0.5 to -2.8°C. In the super-chilling process, the temperature of the food product is reduced to 1-2°C below this range (Duun and Rustad, 2007), which corresponds to 5–30% of the water inside the products forming ice (Wu *et al*., 2014). This temperature is sufficiently low to maintain the taste of products resembling fresh food after thawing. The initial surface freezing results in approximately a 2 mm layer of ice along the product’s surface (Kaale *et al*., 2013), ultimately leading to temperature equilibrium between the exterior and interior during storage (Magnussen *et al*., 2008). This ice formation acts as a refrigeration capacity stored within the product, thereby eliminating the need for external ice.

Martinsdottir (2004) emphasised that using sensory methods to evaluate food provides valuable information on food quality. Martinsdóttir *et al*. (2003) highlighted that freshness is a key element in consumers' assessment of fish quality. Fish are perishable with a limited storage life and are highly dependent on various factors during handling and storage from catch to consumer. Defoe (2014) also discussed the complexity of freshness, noting its varied interpretations, from wild-caught to unfrozen or unprocessed, and even "fresh frozen". Defoe (2014) further stated that quality is linked to freshness, with freshness being essential for quality but not an inherent quality factor.

The shelf life of food refers to the maximum duration for which a specific product remains suitable for human consumption (Tamarit, 2018). For fish, this period extends from the time of catch until it becomes fit for human consumption (Huss, 1995). Various factors influence fish quality, including environmental conditions (such as fishing grounds and seasons), fishing practices, handling and storage conditions, and bleeding and gutting procedures (Dowlati *et al*., 2013). Additionally, the method of catching, overall handling, and fish processing affects fish freshness (Massaquoi, 2011).

Doyle (1989) highlighted that the shelf life of seafood is primarily determined by temperature, which controls the rate of bacterial spoilage, enzymatic activity, and fat oxidation reactions. The spoilage rate decreases at lower temperatures, extending the period of human consumption. Thermometer data loggers are commonly used to monitor temperature fluctuations during shelf life studies (Tamarit, 2018). In normal aerobic chilled storage, the shelf life of fish and fish products is limited by the growth and biochemical activities of specific aerobic spoilage organisms, primarily bacteria. However, cooling to temperatures close to melting ice (0 °C) significantly extends the shelf life of fish and fish products by slowing enzymatic and microbiological activity.

Research studies have investigated the shelf life and storage temperatures of various fish species. According to Magnússon, *et al*., (2010), the freshness declines of whole fish stored in ice depending on the fish species and ambient temperature. The characteristic flavour of fish typically develops after catch and during the initial days of storage, while changes in appearance and texture occur later during storage (Church, 1998). The extent of these sensory changes varies considerably depending on the species and storage methods (Huss, 1995; Church, 1998).

Rigor mortis is a significant change that occurs after the death of the fish. Cooking fish in the pre-rigor stage results in a soft and pasty texture, whereas cooking in the rigor stage leads to a tough but not dry texture. Post-rigor stage cooking results in firm, succulent, and elastic flesh (Huss, 1995).

Seabra and Pedrosa (2010) mentioned that certain compounds, such as astaxanthin found in Atlantic salmon flesh, can influence the shelf life of the product. Astaxanthin, a red carotenoid pigment, possesses significant antioxidant properties that mitigate the detrimental effects of oxidation in food products. This compound occurs naturally in various seafood, including salmon, which obtain it through their diet of zooplankton, insects, or crustaceans that have ingested microalgae that produce astaxanthin. Research indicates that astaxanthin effectively reduces lipid peroxidation while preserving the membrane structure. Additionally, it has beneficial effects in mitigating chronic diseases, cardiovascular diseases, muscular degeneration, and cancer (Seabra and Pedrosa, 2010). Consequently, the addition of astaxanthin could further prolong the shelf life of the product.

According to Coultate (2016), lipids represent a challenging group to categorise, linked by their insolubility in water yet solubility in non-polar solvents like chloroform. Fatty acids, a crucial component of many lipids, significantly contribute to the properties of food owing to their structure. They serve as vital energy sources for most animals, particularly fish. Lipid oxidation, also known as oxidative rancidity, occurs when oxygen interacts with unsaturated fatty acids in an auto-oxidation reaction, resulting in the formation of several chemical products associated with product degradation. This reaction is highly undesirable and prevalent in foods rich in polyunsaturated fatty acids (PUFA) because the α-methylene groups (CH_2-groups adjacent to the double bond) are susceptible to oxidation.

Council (1985) revealed that the microbes responsible for the deterioration of a food product greatly depend on the initial flora of the food and the processing methods applied. According to Forget (2010), the flora of salmon typically consists of psychrotrophic aerobic or facultative anaerobic bacteria. However, the composition of this flora may change during storage. The bacterial growth curve encompasses four primary phases: lag, logarithmic, maximum stationary, and decline. The lag phase may be extended by various processing methods, conservation techniques, and storage conditions, consequently enhancing the shelf life of the product (Forget, 2010).

Drip loss is a critical factor in industrial approaches because it is undesirable and leads to economic losses by reducing product weight (Kiessling, 2004). Additionally, it results in nutrient loss (Dunn, 2008; Turan, 2003). As water leaks out, various compounds exit with it, including sarcoplasmic, water-soluble proteins (Devahastin, 2010). The pH and ice crystal formation significantly influence drip loss, as most of the bulk water shelves within the myofibrillar network. Larger ice crystals can break down more muscle tissue, leading to leakage as the network collapses (Devahastin, 2010). A decrease in muscle pH creates an acidic environment, triggering the release of lysosomal cathepsin, which further breaks down muscle tissue. Consequently, the network holding the water collapses, resulting in water release as drip loss. The structure of muscle tissue fractures from ice crystal formation and breakdown of myofibrillar proteins, causing cell contents to leak out along with leaking extracellular fluid, contributing to drip loss. Drip loss, such as that occurring during freezing/thawing, is undesirable for both consumers and producers, as it not only leads to economic losses but also facilitates faster microbial growth of the thawed product. Many nutrients are water-soluble and exit fish tissue in drip loss, serving as food for microorganisms, which proliferate faster owing to easy access to oxygen. This accelerated microbial growth shortens the shelf life of the product. Moreover, as water freezes, it damages cell structures and increases the solute concentration of the remaining water, resulting in more available water lost as drip during thawing (Devahastin, 2010).

Proteins can undergo various changes, such as oxidation reactions, freeze denaturation, or interactions with lipid oxidation products, all of which tend to render proteins insoluble (Lushchark, 2007). Additionally, freezing a product can induce freeze denaturation of proteins due to several stresses, including ice formation, changes in solute concentration resulting from water crystallisation, and subsequent pH alterations (Lushchark, 2007). Moreover, the oxidation of myofibrillar proteins increases during postmortem muscle storage (Martinaud *et al*., 1997). Myofibrillar proteins constitute approximately 66-77% of the total muscle proteins (Ge *et al*., 2020). While these proteins are insoluble in water, they are soluble in solutions with high salt concentrations (>0.3M). Furthermore, Bhat (2018) explained that oxidation in postmortem muscle also affects protein activity by either inactivating or modifying calpain activity, which is an intracellular enzyme system.

The peroxide value (PV) is a crucial measure for assessing the decomposition status of flesh products, particularly in fish. The formation of peroxides, quantified by PV, is regarded as an indicator of the rate of primary oxidation, whereas the thiobarbituric acid (TBA) value indicates secondary oxidation (Milijasovic *et al*., 2017). Low PV values suggest minimal amounts of primary oxidation products, indicating either low oxidation levels or the breakdown of primary oxidation products into secondary products. Peroxides act as intermediates in this process, and the PV value reflects the disparity between the formation and decomposition of peroxides (Velasco *et al*., 2010). Primary oxidation products have the potential to decompose into various volatile and non-volatile compounds. The rate of primary oxidation product decomposition can outpace their formation, leading to low PV values despite a higher degree of lipid oxidation (Matthäus and Rubner, 2010).

During storage, there is a delay before the primary oxidation products accumulate, as measured via PV titration. The rate of primary oxidation buildup is expected to rise after a few days of storage, followed by a decline as primary oxidation products transform into secondary oxidation products. Consequently, an initial increase in PV values is anticipated, followed by a decrease as secondary oxidation products accumulate.

2.4 Method to evaluate fish quality

2.4.1 Sensory evaluation

Sensory evaluation is the principal method for assessing seafood freshness. It involves the analysis of food sensory characteristics using the human senses. In the fisheries sector, sensory evaluation provides valuable information on fish freshness and quality based on parameters such as appearance, odour, flavour, and texture. Subjective methods rely on assessors' preferences and product acceptability, which can introduce bias among the assessors. This method is commonly used in market research and product development (Huss, 1995).

In fish sensory evaluation, objective descriptive methods are employed using a group of trained assessors (Martinsdottir *et al*., 2009). In this method, structured scales describing the nature and intensity of the quality parameters are used for evaluation. Various grading schemes have been established for the sensory evaluation of whole and cooked fish. The European Union (EU) schemes and Quality Index Methods (QIM) are utilised for evaluating the freshness of raw whole and gutted fish, while the Torry scheme and Quantitative Descriptive Analysis (QDA) are employed for evaluating cooked fish (Martinsdottir *et al*., 2001).

2.4.1.1 Quality Index Method

Huss (1995) mentioned the following in an FAO publication: QIM is based on significant sensory parameters for raw fish, utilising multiple parameters and a scoring system ranging from 0–4 demerit points. QIM employs a practical rating system in which fish are inspected and appropriate demerit points are recorded. The scores for all characteristics are then summed to give an overall sensory score, known as the quality index. The QIM assigns scores of zero for very fresh fish, with larger totals indicating greater deterioration as the fish age.

The most recommended and widely published method is the European Union (EU) scheme, which includes the Quality Index Method (QIM) and Torry schemes (Martinsdottir, *et al*., 2001). The QIM Eurofish, 2001, published by Martinsdottir *et al*. (2001), is considered an ideal tool for determining the quality of fish freshness. Different fish species exhibit various sensory

changes during their spoilage. Therefore, the QIM in Eurofish is based on well-defined characteristic changes that occur in raw fish (Martinsdottir *et al*., 2001).

Scores ranging from 0 to 1, 0 to 2, and 0 to 3 are assigned for changes observed in the odour, texture, and external appearance of the gills, skin, and eyes, as outlined in the grading scheme. A score of zero indicates very fresh fish, with a significant increase in the score as freshness declines. The Quality Index (QI) provides an overall sensory score summarising the scores for all assessed characteristics. Subsequently, QIM schemes have been developed for various fish species, including bogue (*Boops boops*) (Bogdanović et al., 2012) and blackspot seabream (*Pagellus bogaraveo*) (Sants Ana *et al*., 2011). QIM schemes have also been developed for raw fish fillets because of the specific sensory changes observed in different fish species. Bonilla, *et al*. (2007) established a QIM scheme for fresh cod (*Gadus morhua*) fillets based on species-specific characteristics such as colour, odour, texture, gaping, and appearance.

2.4.1.2 Descriptive analysis

According to Huss (1995), descriptive testing is a simple method used to assess a single attribute, such as texture, flavour, or appearance. This method, known as descriptive analysis, can generate a comprehensive description of fish products and has been developed by scientists. This provides a quantitative description of products based on the perceptions of a group of qualified panellists (Defoe, 2017). According to Stone and Sidel (2004), the descriptive test is a very dynamic system in which the panel leader must make numerous decisions when organising a panel through screening, training, and product evaluation. Without sufficient knowledge, inadequate or incorrect product decisions are made. A basic strength of the GDA method is the ability to independently verify (after each test) that individuals perceive differences among products on attributes in a reliable manner. This is directly measured using one-way analysis of variance for each panellist for each attribute. The need to monitor the performance of each panellist in each test reflects the awareness of the sensory limitations of humans (Stone & Sidel, 2004). Flavour profiling is an excellent approach for describing products. The freshness of both whole fish and fillets can be determined through the sensory evaluation of cooked flesh. Sensory evaluation of cooked fish fillets involves the assessment of appearance, flavour, odour, and texture by a sensory panel. A descriptive scale with 10 points has been developed for lean, medium-fat, and fatty fish species, known as the Torry method. A score of 10 is assigned to fish with a very fresh taste and odour, while a score of 3 indicates spoilt fish, and an average score of 5.5 suggests that the fish is nearing its limit for human consumption due to the detection of off-flavour and odour attributes associated with spoilage (Martinsdottir, *et al*., 2001). Scores below 3 are considered unfit for human consumption. Therefore, it is possible to determine the maximum storage time of fish through the sensory evaluation of cooked fish samples. In addition to providing a detailed description of the sensory profile of a product, the maximum storage time of fish can be determined using GDA (Sveinsdottir, *et al*., 2002).

2.4.2 Microbial analysis

Microorganisms are typically found on externally exposed areas of live and newly caught fish, such as the skin, gills, and intestines (Huss, 1998; Bataringaya, 2007). The microbial profile of fish after capture depends on environmental conditions and the microbial quality of water (Shewan, 1977). Other factors influencing microbial growth include temperature, salt content, and natural bacterial flora (Feldhusen, 2000). Fish caught in very cold and clean water environments tend to have lower bacterial counts than those caught in warm water environments (tropical regions). When fish die, bacteria multiply, initially on the skin, and during storage, they invade the flesh of the fish (Huss, 1998; Bataringaya, 2007). Sveinsdottir

et al. (2002) revealed that the number of bacteria on newly caught fish skin varied from 10^2 to 10^7 CFU/cm^2. The TVC on newly caught fish skin was 10^3 CFU/cm^2. Additionally, newly slaughtered salmon contained a total viable count (TVC) of approximately 10 CFU/g in the flesh. If the flesh of healthy, live, newly caught fish is sterile, it is due to the fish's immune system. However, upon death, the immune system collapsed, leading to the termination of the microbial lag phase and the potential onset of exponential growth if not preserved using appropriate preservation methods.

Spoilage of marine temperate water fish is characterised by the development of offensive, fishy, rotten, and H_2S odours and flavours. This differs for different types of fish; tropical and freshwater fish are more likely to develop fruity and sulfhydryl odours and flavours (Gram and Huss, 1996). Microbial examination of fish evaluates the potential presence of bacteria or organisms that may pose a public health risk. It also serves as an indication of the hygiene and quality of fish, reflecting factors such as temperature abuse and hygiene conditions during handling (Lauzon, *et al*., 2010). The number of specific spoilage bacteria provides information on the remaining shelf life, with higher counts indicating a more advanced stage of spoilage (Huss, 1995). For microbiological assessments, it is recommended to use measurements of specific spoilage organisms or Total Viable Counts (TVC) (Olafsdottir, *et al*., 1997). Various peptone-rich substrates containing ferric citrate have been employed for the detection of H_2S-producing bacteria, such as *Shewanella putrefaciens*, which form black colonies due to the precipitation of Ferrous Sulphide (FeS) (Huss, 1995). In aerobic storage conditions, levels of 10^8-10^9 CFU/g of specific spoilage bacteria in the flesh are required to cause spoilage during ice storage (Gram and Huss, 1996). The ice-chilled salmon was microbiologically rejected when compared to the 10^5 CFU/g (log CFU/g = 5.0) threshold in the flesh of whole salmon, which led to rejection at 20 days, as reported in Sveinsdottir, *et al*. (2002).

2.4.3 Chemical analysis

The chemical composition of fish is another important factor that directly affects fish quality and sensory characteristics (Huss, 1988). Autolytic changes are responsible for the initial loss of quality in fresh fish, but their contribution to the spoilage of chilled fish and fish products is minor. Autolytic changes are of great importance in frozen fish (Huss, 1995). Chemical compounds developed in naturally spoiling fish and sterile fish indicate that most volatile compounds are produced by bacteria (Shewan, 1962). Typically, many specific spoilage bacteria on fish can use Trimethylamine N-Oxide (TMOA) as an electron acceptor in anaerobic respiration. The reduced chemical compound trimethylamine (TMA), which is one of the leading components of spoilt fish, generates a fishy odour (Dalgaard *et al*., 1993). Bacterial activity is inhibited, and TMOA is broken down by autolytic enzymes into dimethylamine (DMA) and formaldehyde (FA). The formation of formaldehyde causes increased denaturation of fish tissue, changes in texture, and loss of water retention capacity (Huss, 1995). Fat oxidation usually occurs after autolysis and bacterial spoilage when fresh fish are iced. The formation of hydroperoxides is the first step in the oxidation process, and although tasteless, it causes brown and yellow discoloration of fish tissue (Huss, 1995).

The chemical method involves analyzing a sample to determine the concentration of a specific chemical (Howgate, 1982). Various chemical methods are used to measure total volatile bases (TVB) in fish, but with each change, the fish or fish extract is made alkaline. Owing to the chemical change, the generated bases are distilled off, collected, and measured by titration. An established method for determining TVB-N is based on the water stream distillation of a perchloric acid extract (Oehlenschlager, 1997). According to European Commission Regulation No. 2074 (2005), unprocessed fishery products are regarded as unfit for human consumption when sensory assessment raises doubts regarding freshness and chemical analysis reveals that

TVB-N has exceeded certain limits for specific fish species, such as 25 mgN/100 g maximum for perch species, 30 mgN/100 g for flatfish, and 35 mgN/100 g for white lean fish (cod and haddock).

3 METHODOLOGY

3.1 Experimental design

The experiment was conducted at Matis laboratories in Reykjavik, Iceland. The experimental design is illustrated in **Error! Reference source not found.**.

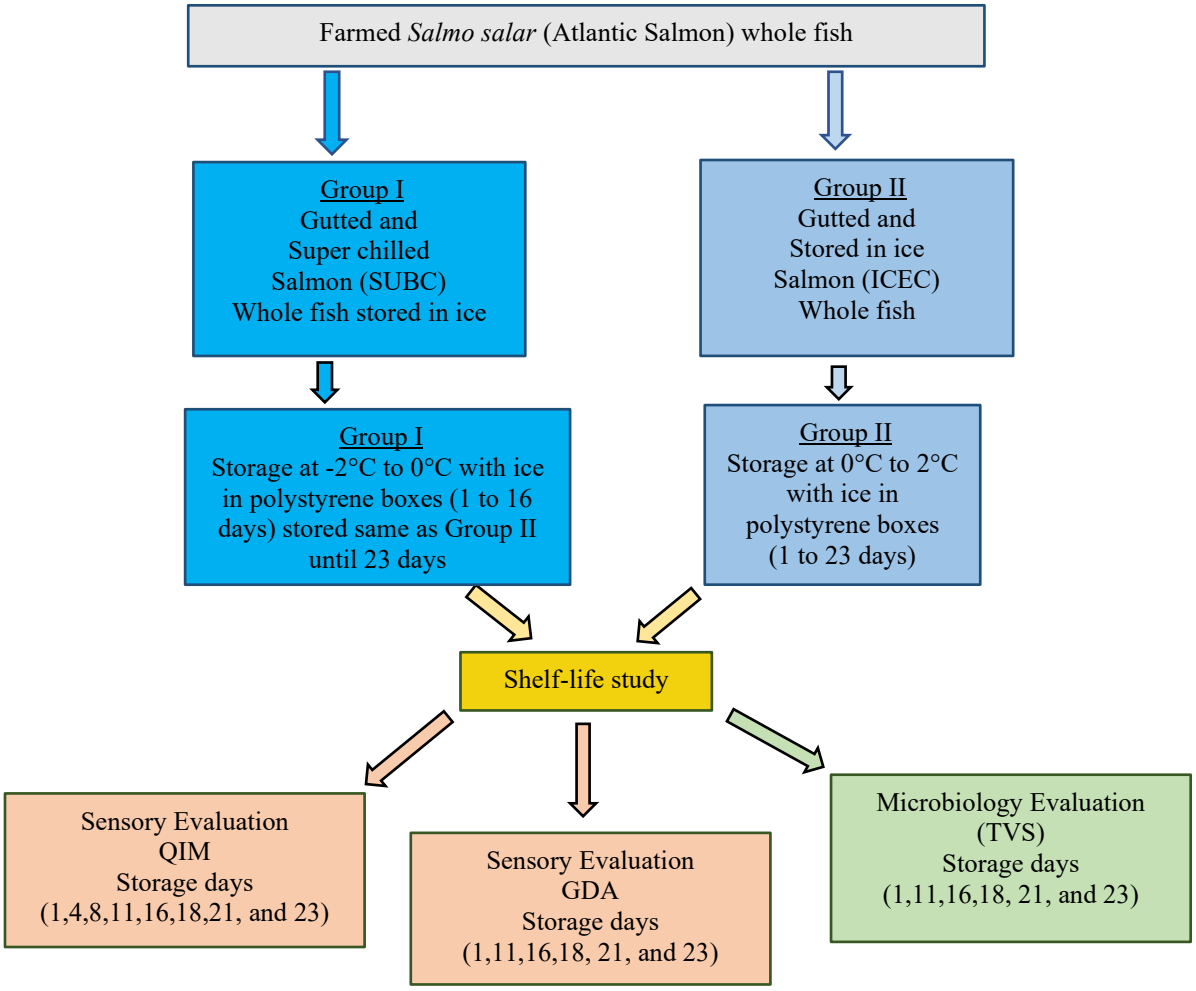


Figure 1. Flow chart for the experimental design for shelf-life study of super-chilled and ice chilled farmed Salmo salar (salmon) whole fish storage at 00C to 40C with crushed ice in polystyrene boxes.

Group I, SUBC salmon, was transferred to a 0–2°C storage room on day 16, as a storage temperature of -2°C was utilised during sea transportation, which typically takes around 10 to 14 days. Three fish from both ICEC and SUBC salmon were taken out each day (1, 4, 8, 11, 14, 16, 18, 21, and 23), and microbial analysis and sensory evaluation (Quality Index Method - QIM and Generic Descriptive Analysis – GDA) were conducted for the shelf-life study in ice storage according to Table 1.

Table 1. Sampling schedule during the study period.

Experiment date	No of dates in ice storage	Microbiological Analysis	Sensory evaluation	
			QIM Evaluation	GDA Analysis
05.03.2024	01	X	X	X
08.03.2024	04		X	
12.03.2024	08		X	
15.03.2024	11	X	X	X
18.03.2024	14		X	
20.03.2024	16	X	X	X
22.03.2024	18	X	X	X
25,03,2024	21	X	X	X
27.03.2024	23	X	X	X

3.2 Raw material

The cultured amphihaline fish species *Salmo salar* Linnaeus, 1758 (Atlantic salmon) was gutted, and whole fish were used for the study. The raw materials were provided free of charge by Arnarlax, one of the biggest producers of cultured salmon in Iceland. A total of 54 salmon fish (4.24 ± 0.73 kg) were provided by Arnarlax within 1 d of slaughter. Half of the fish (n=27) were pretreated using the super-chilling technique and then packed in polystyrene boxes with ice (SUBC). The remaining 27 fish were cooled with ice and packed in polystyrene boxes with ice (ICEC). Both ICEC and SUBC boxes included three salmon fish each and were transported to Matis for the study. The SUBC salmon boxes were stored in a -2 °C refrigerated cool room for 16 days before being transferred to the 0ºC to 4 °C cool room for the remainder of the experimental period. Conversely, the ICEC salmon boxes were stored in 0ºC–4 °C cool rooms throughout the entire 23-day experimental period.

3.3 Temperature monitoring

Button temperature loggers (DS1922L/T) were used to measure and record the temperature every 15 min during the ice storage period. Arnarlax staff placed eight button temperature loggers in one selected ICEC and SUBC box as follows: two temperature loggers were placed on the outside of the polystyrene box, another two were placed on the wall inside the box, two were placed on the fish, and the remaining two were inserted into the flesh of the fish, closer to the back fin. All temperature data loggers were installed on 4 March 2024 and recovered on 23 March 2024 at the end of the shelf-life study of ICEC and SUBC cultured Atlantic salmon stored in ice.

3.4 Sensory evaluation

3.4.1 QIM

Three fish from each group (ICEC and SUBC) were placed on a clean white table for 45 min before the QIM evaluation. Each fish sample was coded with a three-digit random number. All physical observations were conducted under standardised conditions at room temperature and under white fluorescent light. For the fish, the side from which samples were taken for microbiology faced down, as shown in Figure 2.

Figure 2. Salmon sample arrangement for QIM sensory evaluation.

The QIM scheme for cultured Atlantic salmon (*Salmo salar),* as explained in the QIM Eurofish Sensory Guideline (Martinsdóttir *et al*., 2001), was utilised for the alteration of the QIM scheme for the same species stored with chilled ice in polystyrene boxes after the SUBC pretreatment and ICEC conditions. A detailed description of both groups was completed during the ice storage period, representing dates 1, 4, 8, 11, 14, 16, 18, 21, and 23 (end of the expected shelf life). The QIM assessments were conducted by trained panellists at Matis and by 2-4 GRO-FTP fellows (who were trained in QIM sensory evaluation of cod). The Quality Index (QI) was obtained following the quality index method scheme for farmed Atlantic salmon, as indicated in Appendix A.

3.4.2 GDA

For the GDA analysis, whole salmon from both the ICEC and SUBC groups were filleted after the QIM sessions, and samples weighing approximately 40-50 g were taken from the loin part of the fillets. The samples were placed in aluminium boxes coded with three-digit random numbers. Subsequently, the samples were cooked at 100°C for seven minutes in a pre-warmed Convostar oven with air circulation and steam (Figure 3).

Figure 3. Preparation of cooked samples for GDA sensory evaluation in Matis.

Generic descriptive analysis (GDA), as introduced by Stone and Sidel (2004), was utilised to evaluate cooked samples of salmon in two groups: ICEC and SUBC. An unstructured scale ranging from 0 to 100% was used to describe specific attributes such as odour, flavour,

appearance, and texture. However, in this study, the GDA panellists focused solely on the odour and flavour (Table 2). The sensory evaluation of the cooked salmon was conducted as shown in Table 3. The panellists observed differences in the odour and flavour of the cooked salmon, with replicates of both ICEC and SUBC provided in the same area of the fillet. Red light was used to mask the appearance during evaluation.

Table 2. Sensory attributes (odour and flavour) evaluated according to Generic Descriptive Analysis for cooked salmon

	Sensory attribute	Short name	Description of attribute
	Odour		
01	Sweet, characteristic	O-sweet	Sweet characteristic odour of boiled salmon
02	Fresh oil	O-oil	Fresh unspoiled fish oil
03	Spoilage sour	O-sour	Spoilage sour, spoilage characteristic
04	Rancid	O-rancid	Rancid odour, spoilage characteristic
05	Queasy sweet	O-queasy	Queasy sweet, spoilage characteristic
06	Putrid	O-putrid	Putrid odour
07	Spoilage odour	O-spoilage	Overall spoilage odour
	Flavour		
08	Sweet, characteristic	F-sweet	Sweet characteristic flavor of boiled salmon
09	Fresh oil	F-oil	Fresh fish oil
10	Spoilage sour	F-sour	Spoilage sour, spoilage characteristic
11	Rancid	F-rancid	Rancid flavour, spoilage characteristic
12	Queasy sweet	F-queasy	Queasy sweet, spoilage characteristic
13	Spoilage flavour	F-spoilage	Overall spoilage flavour

Twelve panellists participated in the GDA. Both ICEC and SUBC cooked salmon samples were evaluated by four to eight trained panellists in each session. The panellist's participation is indicated in Table 3, and only one panellist participated in all the GDA sessions. Maintaining continuity in the panellists' participation was a challenging task due to their scheduled work and the Easter vacation.

Table 3. The panellist's participation to the GDA evaluation

Panellists	**Ice storage period/Days**					
	01	**11**	**16**	**18**	**21**	**23**
01	X	X	X	X	X	X
02		X	X	X	X	X
03	X		X	X	X	
04	X					
05	X					
06					X	X
07			X	X	X	X
08		X	X			
09	X	X		X		
10	X	X	X			
11		X	X			
12	X			X	X	X
Total # each day	07	06	08	06	06	05

All panellists had been trained according to international standards (EN ISO 8586:2014), including the detection and recognition of tastes and odours, the use of scales, and the development and use of descriptors. Each panellist evaluated duplicates of each test group in a random order (four samples per session). Data collection was facilitated using a computerised system (FIZZ, Version 2.51C, 1994–2014, Biosystèmes).

3.5 Microbial analysis

Flesh samples from both salmon groups (ICEC and SUBC) were collected for microbiological analyses. Samples were collected from fresh salmon after different ice storage periods (1, 11, 16, 18, 21, and 23 days) before initiating sensory evaluation. Total Viable Counts (TVC) were determined on molten nutrient agar using the pour plate method. Each sample (20 g) was minced using a laboratory-grade blender. The minced sample was then diluted with 200 ml of cooled Maximum Recovery Diluent (MRD, Oxoid, UK) and homogenised in a stomacher bag for one minute. Serial 10-fold dilutions were prepared using 9 ml of cooled MRB. After completing the dilutions, the IA plate solution was spread onto the plates, which were then incubated at 17°C for 5 days. The total number of colonies was counted using a colony counter, and the total viable bacterial count was calculated as colony-forming units (CFU) per gram (g) of the sample. Each colony on the plate represented one bacterium in the fish tissue.

3.6 Data analysis

Microsoft Excel Professional Plus 2016, which is part of the Office 365 software package, was used for various analytical tasks. It was employed to compute the means for parameters such as Total Viable Count (TVC), Quality Index Method (QIM), and other necessary calculations. Additionally, it was used to create plots against storage time. Furthermore, Excel facilitated the calculation of the linear regression and correlation equations for the QIM analysis. To analyse whether the ICEC and SUBC groups differed in QI scores with respect to ice storage time, an ANOVA was conducted to compare the p-values using R (version 4.3.3 for Windows). Each panellist's performance was evaluated using the Panelcheck V1.4.0 (Nofima, Tromsø, Norway) software package. Statistical analysis of the results was conducted using the NCSS 2000 (NCSS, Utah, USA) software package and Microsoft Excel 2016. Analysis of variance (ANOVA) using the General Linear Model method and Duncan´s post hoc test were employed to perform multiple comparisons on GDA data. The data for each sampling day were analysed as separate datasets. A significance level of $p < 0.05$ was applied in the ANOVA; values below this threshold indicated significant differences between the samples of each group (ICEC and SUBC).

4 RESULTS

4.1 Temperature fluctuation in storage

The experiment commenced on 5 March 2024 and concluded on March 27th, 2024 (1-23 days period). Subchilled (SUBC) salmon boxes were stored in a cool room at -2°C to 0°C, whereas ice-chilled (ICEC) salmon boxes were stored in a cool room at 0°C to 4°C. Four button temperature loggers were placed in one selected box from each group, as listed in Table 4. Eight button temperature loggers were recovered from the boxes, and the data were provided as an Excel file. One data logger, which was placed on ice inside the ICEC box, failed to obtain readings because of technical issues. Additionally, data from three temperature loggers (No. 3, 7, and 8) were erased due to recent data overwriting previously recorded data. However, the loss of data did not affect the measurement of the overall temperature fluctuations during the study period. The remaining temperature data are presented as a temperature range and average temperature, with the standard deviation calculated using a total of 20,565 temperature data points, as shown in Table 4.

Table 4. The button temperature logger's recovery data

No	Group	Recovered place	Temperature range	Average temperature
01	SUBC	Core of the one fish	-0.98 to -0.40 ºC	-0.94 ºC ± 0.09
02		Inside the box, on ice	-2.50 to -0.05 ºC	-1.86 ºC ± 0.59
03		Inside the box, fixed on the wall	Data erased	
04		Outside the box, fixed on the wall	-2.50 to -0.05 ºC	-0.85 ºC ± 0.78
05	ICEC	Inside the box, on ice	Technical Issue	
06		Core of the one fish	0.04 to 0.54 ºC	0.40 ºC ± 0.25
07		Inside the box, fixed on the wall	Data erased	
08		Outside the box, fixed on the wall	Data erased	

The results showed that the temperature loggers inserted in the core of the fish (approximately 30 mm beneath the flesh, next to the back fin of the fish) recorded an average temperature of -0.9°C ± 0.1 for SUBC salmon and 0.4°C ± 0.3 for ICEC salmon during the ice storage period. The temperature range (core of fish) also varied, with SUBC salmon ranging from -1.0 to -0.4°C and ICEC salmon ranging from 0.0 to 0.5°C. According to the Arnarlax report provided with the salmon samples, the indicated average core temperature for SUBC salmon was -0.4°C ± 0.0, and for ICEC salmon, it was 1.9°C ± 0.1. However, during the experimental period, the core temperature logger readings showed an average of 0.5°C higher for SUBC salmon and 1.5°C lower for ICEC salmon than the reported values. Additionally, for SUBC salmon boxes on ice, the temperature range and average temperature were -2.5°C to -0.1°C and -1.9°C ± 0.6 °C, respectively. The temperature logger placed outside the SUBC salmon box recorded a mean temperature inside the refrigerated cool room with a range of -2.5°C to -0.1°C and an average value of -0.9°C ± 0.8 during the study period (Table 4).

4.2 Sensory evaluation and shelf-life study in ice storage

4.2.1 Sensory evaluation using QIM scheme

Fifteen assessors participated in the QIM sessions, as shown in Table 5. Two assessors (Assessors 11 and 12) participated in only one session, and their QI values, along with those of two other assessors (Assessors 05 and 07), were not included in the results interpretation due to their below-average and above-average values, respectively. Maintaining continuity in the

panellists' participation was challenging due to their busy work schedules and the Easter vacation.

Table 5. The assessors' participation in the QIM sessions each day.

Date	Days	Assessors														
		01	02	03	04	05	06	07	08	09	10	11	12	13	14	15
05.03.2024	01	X		X	X	X	X	X	X	X					X	X
08.03.2024	04	X	X	X	X	X	X	X	X				X	X	X	X
12.03.2024	08	X	X			X	X	X	X			X		X	X	X
15.03.2024	11	X		X	X			X	X	X					X	X
18.03.2024	14	X			X	X	X		X						X	X
20.03.2024	16	X	X		X				X		X				X	X
22.03.2024	18		X			X	X		X		X				X	X
25.03.2024	21		X			X	X								X	X
27.03.2024	23		X			X	X				X				X	X

The quality index (QI) was calculated based on the sum of the scores of the panellists for each storage day for both ice-chilled (ICEC) and sub-chilled (SUBC) pre-treated cultured salmon during the ice storage period. Figure 4 depicts the distribution of the QI scores over time for the ICEC salmon in ice storage for each assessor. Some differences were observed between the assessors, especially at the end of the experiment, except on day 23.

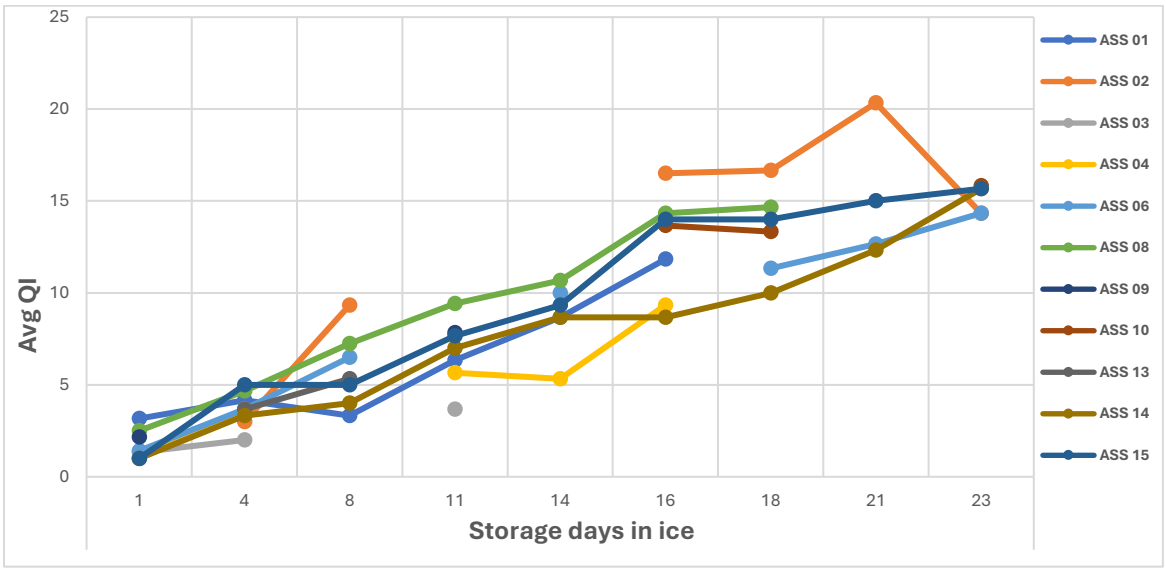


Figure 4. Assessors' average QI for ICEC salmon during iced storage.

Figure 5 illustrates the distribution of the QI scores over time for SUBC salmon in ice storage between the assessors. Similar trends in assessor differences were observed in the ICEC group.

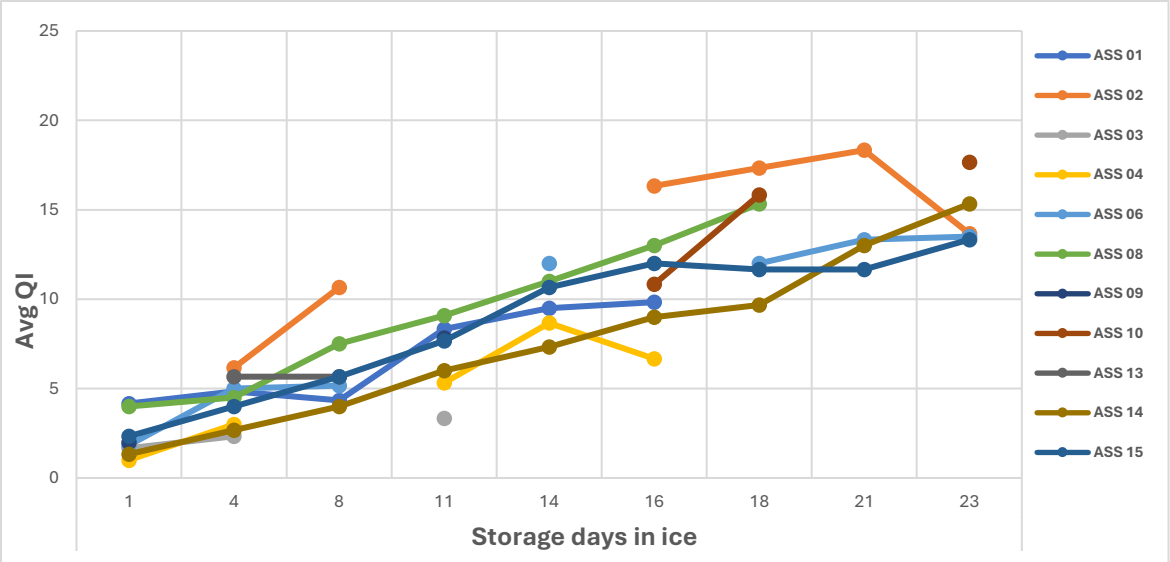


Figure 5. Assessors' average QI for SUBC salmon during iced storage.

The variation increased with storage time, indicating that the panellists were more consistent in their assessments when analysing very fresh salmon with the QIM scheme at the beginning of the storage period than with less fresh salmon at a later stage in both the SUBC and ICEC groups (Figures 4 and 5). There was a tendency for some panellists to score either higher or lower than the average score obtained throughout the storage period. The evaluated sum of scores according to the QIM scheme for farmed Atlantic Salmon is presented as the QI. The average QI and p-values for each group of ICE and SUBC salmon are presented in Table 6. The average QI was calculated for both groups of salmon (ICEC and SUBC) on each storage day of sampling and formed a linear relationship with the ice storage time (Figure 6). A high correlation between the average QI and days in ice storage was observed, with $R^2 = 0.9717$ for ICEC group and $R^2 = 0.9863$ for the SUBC group (Figure 6).

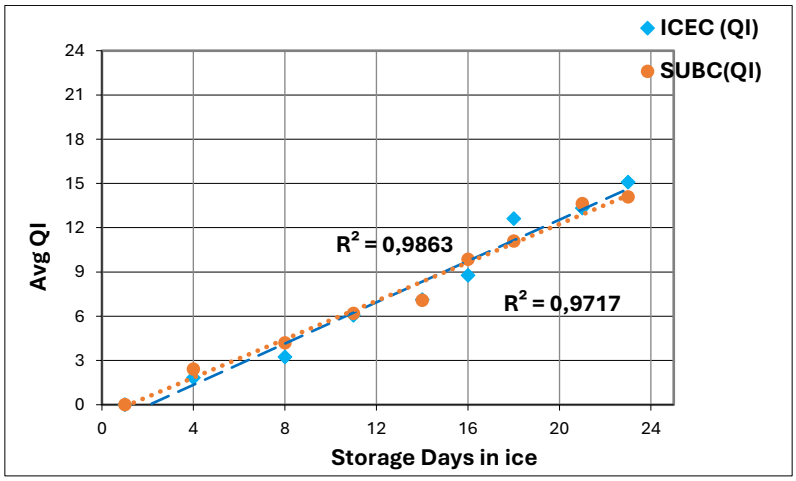


Figure 6. Linear relationship of QI values (ICEC and SUBC groups) and storage time in ice.

According to the ANOVA analysis conducted to compare the ICEC and SUBC groups QI scores with respect to ice storage time, the obtained p-values are indicated in Table 6. Based on the appropriate hypothesis (H_1) that there are no differences between the two groups of ICEC and SUBC salmon with days in ice storage when testing with sensory evaluation parameters, the average QI values were recorded for both ICEC and SUBC salmon groups across different days of ice storage.

Table 6. Average QI scores and p-values for salmon (ICEC and SUBC) storage days in ice.

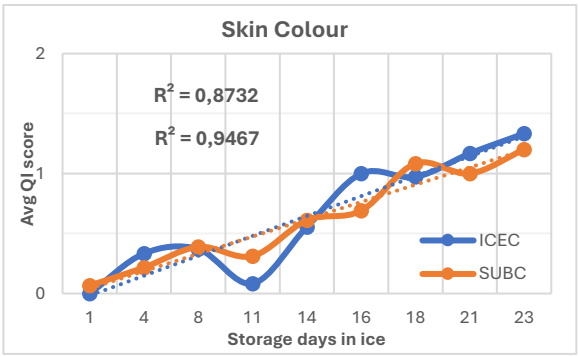
Days in Ice	Average Quality Index (QI)		P- value
	ICEC	SUBC	
1	1.8 ± 0.9	2.4 ± 1.7	0.06
4	3.3 ± 1.2	4.2 ± 1.8	0.22
8	6.1 ± 2.5	6.2 ± 3.0	0.86
11	7.1 ± 2.4	7.1 ± 2.4	0.95
14	8.8 ± 2.2	9.9 ± 2.3	0.16
16	12.6 ± 3.1	11.1 ± 3.3	0.13
18	13.3 ± 2.5	13.6 ± 3.1	0.75
21	15.1 ± 3.6	14.1 ± 2.8	0.46
23	15.2 ± 1.4	14.7 ± 2.8	0.57

According to the obtained p-values, all values were greater than 0.05 ($p > 0.05$) at a 95% confidence level. This indicates that the p-values for the average QI across different ice storage dates were greater than 0.05, suggesting that the deviation from the null hypothesis (H_0) was not significant. In other words, there was no significant difference between the two groups of ICEC and SUBC Atlantic salmon across various storage dates in ice. However, on days 1, 4, and 8, the average QI values for the SUBC group were higher than those for the ICEC group in ice storage. On day 11, both groups had the same average QI value. On day 14, the average QI values for SUBC salmon were greater than those for ICEC salmon. Conversely, on days 16 and 18, the results were reversed for the average QI. In the later stages of ice storage, specifically on days 21 and 23, the average QI values for the SUBC group were higher than those for the ICEC group.

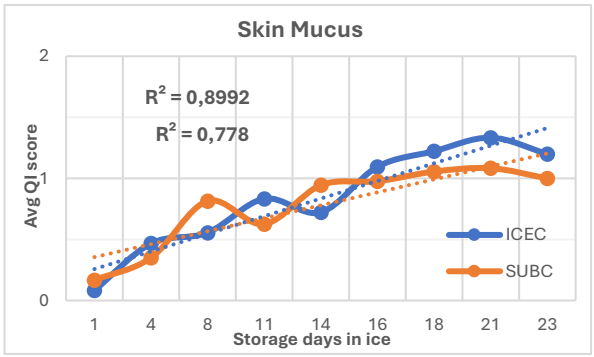
Table 7. Average scores for each quality parameter in QIM scheme and average QI of SUBC and ICEC group's salmon in ice storage period (with linear regression (R^2) and PLC regression (r)values)

#	Quality parameter	Group	Days in ice storage									R^2	PLC (r)
			01	04	08	11	14	16	18	21	23		
1	Skin colour	ICEC	0.0	0.3	0.4	0.1	0.6	1.0	1.0	1.2	1.3	0.87	0.75
		SUBC	0.1	0.2	0.4	0.3	0.6	0.7	1.1	1.0	1.2	0.95	0.78
2	Skin mucus	ICEC	0.1	0.5	0.6	0.8	0.7	1.1	1.2	1.3	1.2	0.89	0.71
		SUBC	0.2	0.4	0.8	0.6	0.9	1.0	1.1	1.1	1.0	0.78	0.59
3	Skin odour	ICEC	0.1	0.3	0.7	0.5	0.4	1.0	0.9	1.2	1.1	0.86	0.69
		SUBC	0.1	0.3	0.4	0.5	0.5	0.8	1.1	1.2	1.2	0.96	0.75
4	Skin texture	ICEC	1.0	1.0	1.1	1.0	1.0	1.1	1.1	1.3	1.0	0.24	0.37
		SUBC	1.0	1.0	1.2	1.0	1.0	1.2	1.3	1.1	1.0	0.08	0.33
5	Eyes pupils	ICEC	0.1	0.2	0.2	0.1	0.4	0.7	1.0	1.4	1.1	0.86	0.72
		SUBC	0.2	0.3	0.4	0.3	0.7	0.7	1.2	1.6	1.5	0.91	0.71
6	Eyes form	ICEC	0.3	0.7	0.8	0.9	1.1	1.5	1.2	1.5	1.5	0.86	0.70
		SUBC	0.4	0.9	0.7	1.0	1.2	1.3	1.6	1.5	1.5	0.89	0.67
7	Gills colour	ICEC	0.1	0.1	0.3	0.9	0.9	1.4	1.6	1.8	1.6	0.92	0.84
		SUBC	0.1	0.2	0.6	0.8	1.6	1.3	1.1	1.5	1.5	0.78	0.74
8	Gills mucus	ICEC	0.1	0.5	0.6	1.4	1.5	1.4	1.6	1.7	1.7	0.84	0.76
		SUBC	0.2	0.5	0.6	1.0	1.6	1.4	1.5	1.7	1.8	0.91	0.76
9	Gills odour	ICEC	0.1	0.2	0.8	1.0	1.3	1.8	1.8	2.1	2.3	0.96	0.88
		SUBC	0.1	0.3	0.6	0.9	1.0	1.4	1.7	1.7	1.8	0.98	0.87
10	Abdomen blood	ICEC	0.0	0.0	0.2	0.1	0.1	0.3	0.4	0.2	0.6	0.71	0.53
		SUBC	0.0	0.0	0.1	0.2	0.1	0.4	0.5	0.3	0.5	0.77	0.63
11	Abdomen odour	ICEC	0.0	0.1	0.5	0.3	0.8	1.2	1.5	1.5	1.7	0.94	0.80
		SUBC	0.2	0.2	0.4	0.5	0.8	1.0	1.5	1.5	1.6	0.95	0.78

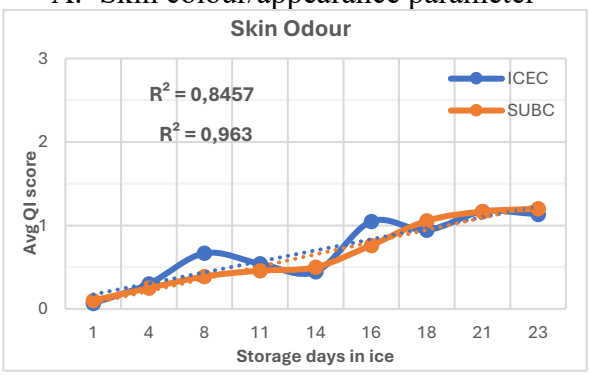
Both the ice-chilled (ICEC) and subchilled (SUBC) pretreated salmon groups in ice storage indicated a clear linear relationship ($R^2 \geq 0.9$) with time in ice storage for most of the quality parameters, except for a few, as shown in Figure 9. However, there was a clear linear relationship for quality parameters such as skin colour/appearance ($R^2 = 0.9467$) (Figure 07: A, skin odour ($R^2 = 0.9630$) (Figure 9: C, eyes pupil appearance ($R^2 = 0.9125$) (Figure 07: E, and gills mucus ($R^2 = 0.9085$) (Figure 07: H, respectively, in the SUBC salmon group. Additionally, there was a clear linear relationship between quality parameters, such as gill colour ($R^2 = 0.9182$) (Figure 07: G) in the ICEC group of salmon. Furthermore, there was a strong linear relationship for gill odour ($R^2 = 0.9797$ and $R^2 = 0.9636$) in Figure 07: I and abdomen odour ($R^2 = 0.9392$ and $R^2 = 0.9518$) in Figure 07: K, for both ICEC and SUBC salmon. The lowest linear regression values were obtained for the skin texture attribute, with $R^2 = 0.02$ for ICEC salmon and $R^2 = 0.02$ and $R^2 = 0.08$ for SUBC salmon.



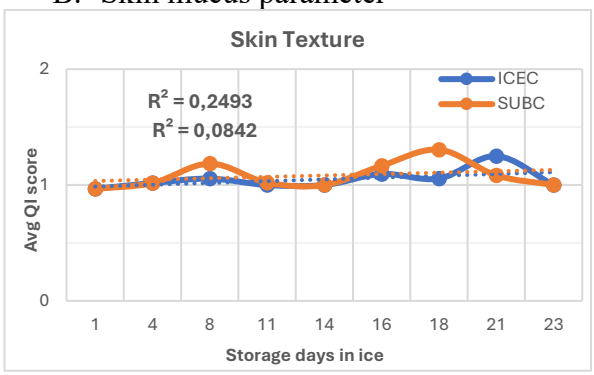
A. Skin colour/appearance parameter



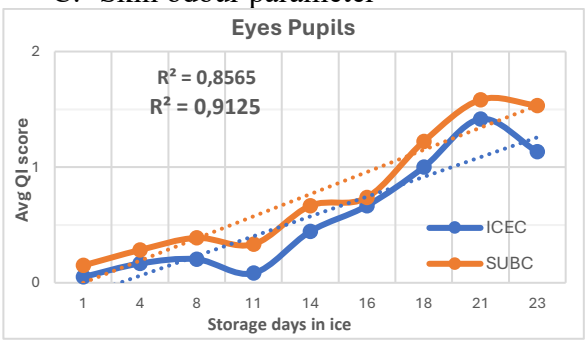
B. Skin mucus parameter



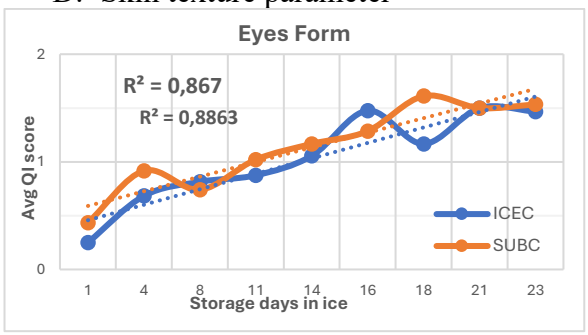
C. Skin odour parameter



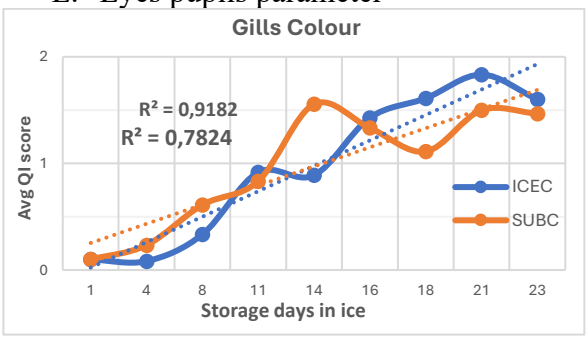
D. Skin texture parameter



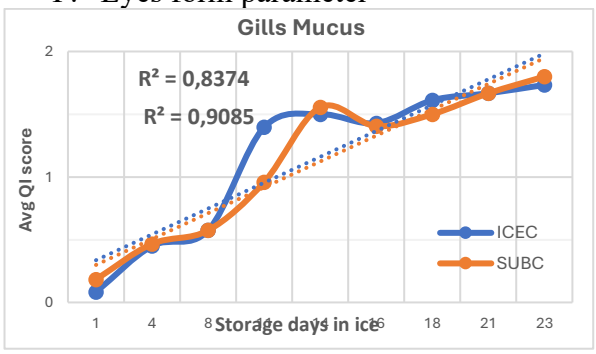
E. Eyes pupils parameter



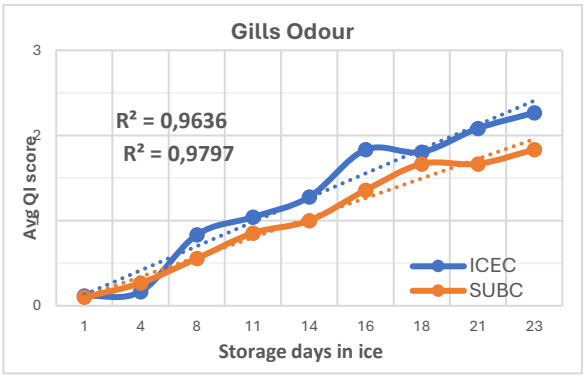
F. Eyes form parameter



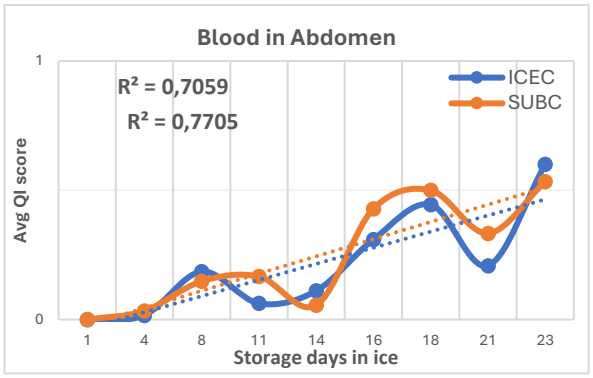
G. Gills colour/appearance parameter



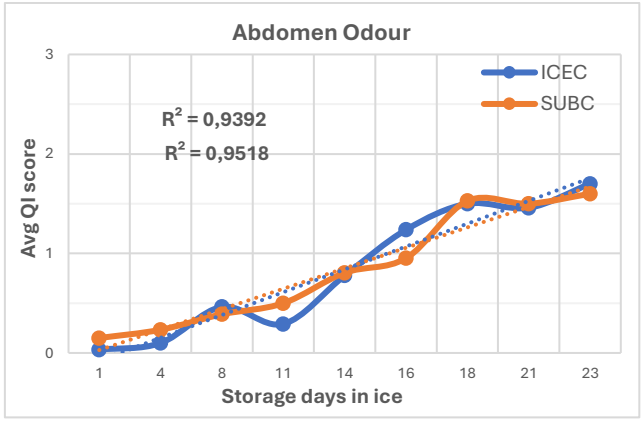
H. Gills mucus parameter



I. Gills odour parameter



J. Blood in abdomen parameter



K. Abdomen odour parameter

Figure 7. *Variation of eleven QI parameters of QIM scheme with ice storage period.*

Photographs taken for visual quality parameters, including skin colour/appearance, eyes pupils, eyes form, gills colour/appearance, and blood in the abdomen, indicated changes in the mentioned quality parameters with the ice storage period. However, the most dominant changes occurred in the eye pupils and gill colour/appearance (Figures 7 and 8). The skin appearance/colour quality parameter in the QIM scheme for days 1, 11, 14, and 23 of ice storage is shown in Figure 8.

	
A. Day 01 ICEC, ice storage	B. Day 01 SUBC, ice storage

C. Day 11 ICEC, ice storage

D. Day 11 SUBC, ice storage

E. Day 14 ICEC, ice storage

F. Day 14 SUBC, ice storage

G. Day 23 ICEC, ice storage

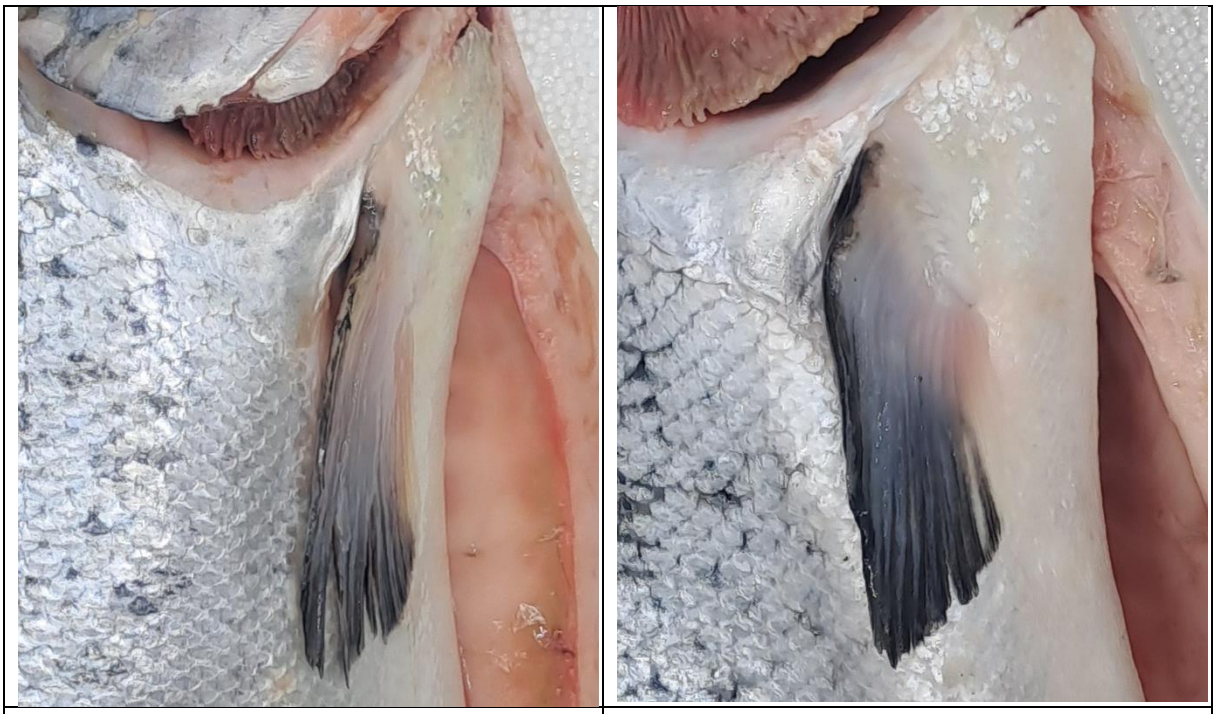
H. Day 23 SUBC, ice storage

Figure 8. Variation in skin colour/appearance of ICEC and SUBC salmon during the ice storage period for selected days.

When considering the skin appearance/colour quality parameter in the QIM scheme, the average scores showed a linear relationship with the dates of ice storage for both the ICEC and SUBC groups (Figure 7 A and Table 7). The skin appearance/colour quality attribute scores were assigned according to the QIM scheme, where a score of 0 represents a shiny pearl appearance all over the body, a score of 1 indicates less pearl shininess, and a maximum score of 2 is given to fish that appear yellowish, especially near the abdomen. On day 1, at the beginning of the experiment, the average score for the skin colour appearance attribute was 0.3, indicating a value closer to zero. Subsequently, on days 8 and 11, the scores remained close to zero for both the ICEC and SUBC groups. On day 14 of ice storage, the same average score (0.6) was recorded for skin colour in both groups of salmon. However, on day 16, the average skin colour score was 1.0 for the ICEC group and 0.7 for the SUBC group. As ice storage continued, on days 18 and 21, the average skin appearance scores exceeded 1. By the end of the experiment on day 23, the average skin appearance scores were 1.3 and 1.2 for the ICEC and SUBC groups of salmon in ice storage, respectively (Figure 7A and Table 7).

A. Day 1 ICEC, Ice storage

B. Day 1 SUBC, Ice storage



C. Day 23 ICEC, Ice storage | D. Day 23 SUBC, Ice storage

Figure 9. Skin mucus quality parameters on days 1 and 23 of the ICEC and SUBC groups of salmon in ice storage.

Skin mucus, the second quality parameter in the QIM scheme, showed average scores that exhibited a linear relationship with the dates of ice storage for both ICEC and SUBC salmon. However, it is not indicated to have a strong linear relationship; nonetheless, the trend is comparatively fair with the dates of ice storage (Figure 7: B and Table 7). Furthermore, the R-squared (R^2) values for skin mucus were 0.899 and 0.788 for the ICEC and SUBC salmon groups, respectively. Skin mucus quality attribute scores were assigned according to the QIM scheme, where a score of 0 represented clear and non-clotted mucus, a score of 1 indicated milky and clotted mucus, and a maximum score of 2 was given to fish that exhibited yellowish and clotted mucus on the skin. The average skin mucus score was 0.1 and 0.2 (considered as 0) for ICEC and SUBC salmon on day 1 of ice storage, respectively. From then until day 16, the average scores for both groups of ICEC and SUBC salmon remained below 1, indicating a clear, not clotted condition. However, after day 16, the average values exceeded 1, indicating a shift towards milky, clotted mucus. On days 16, 18, 21, and 23, the average skin mucus score for ICEC was comparatively higher than that for SUBC. The highest average score of 1.3 was recorded for ICEC salmon on day 21, after which the average scores for skin mucus decreased in both the ICEC and SUBC groups (Figure 7 B and Table 7).

As the third quality attribute in the QIM scheme, skin odour also exhibited average scores indicating a linear relationship with the dates of ice storage for both the ICEC and SUBC salmon. A strong linear relationship was found with the dates of the ice storage period (Figure 9 B and Table 4). The R-squared (R2) values for skin odour were 0.846 and 0.963 for the ICEC and SUBC salmon groups, respectively (Figure 7 C and Table 7). The skin odour quality attribute scores were assigned according to the QIM scheme, where a score of 0 represented a fresh, sea-weedy, or neutral odour, a score of 1 indicated a cucumber, metallic, or hay-like odour, a score of 2 was given to fish with a sour or dishcloth odour, and a maximum score of 3 was allocated to the rotten odour detected on the skin (Appendix 1). The average skin odour

score was almost 0 (0.1 for both) for both ICEC and SUBC salmon on day 1 of ice storage. From then until day 14, the average scores for both the ICEC and SUBC groups remained below 1, indicating a fresh, sea-weedy, or neutral odour from the skin. However, after day 16, the average values exceeded approximately 1, indicating a shift towards a cucumber-, metallic-, or hay-like odour emanating from the skin. On days 18, 21, and 23, the average skin odour score for ICEC salmon was comparable to that of SUBC salmon. The highest average score of 1.2 was recorded for SUBC salmon on day 23 (Figure 7 C and Table 7).

The average skin texture attribute score showed a weak linear relationship with the number of days of ice storage. The R-squared values were $R^2 = 0.249$ and $R^2 = 0.084$ for the ICEC and SUBC groups, respectively. The scoring regime for skin texture was as follows: a score of 0 was given when the skin texture felt rigid when pressed down with fingertips, a score of 1 was assigned when finger marks on the skin disappeared rapidly, and a maximum score of 2 was allocated when the finger mark remained visible for over a 3-second period after pressing on the skin. At the beginning of the experiment on day 1 and at the end of the experiment on day 23, the average scores for skin texture were almost equal to 1, indicating that the finger marks disappeared rapidly.

A. Day 01 ICEC, ice storage

B. Day 01 SUBC, ice storage

C. Day 11 ICEC, ice storage

D. Day 11 SUBC, ice storage

E. Day 14 ICEC, ice storage

F. Day 14 SUBC, ice storage

G. Day 18 ICEC, ice storage

H. Day 18 SUBC, ice storage

I. Day 21 ICEC, ice storage

J. Day 21 SUBC, ice storage

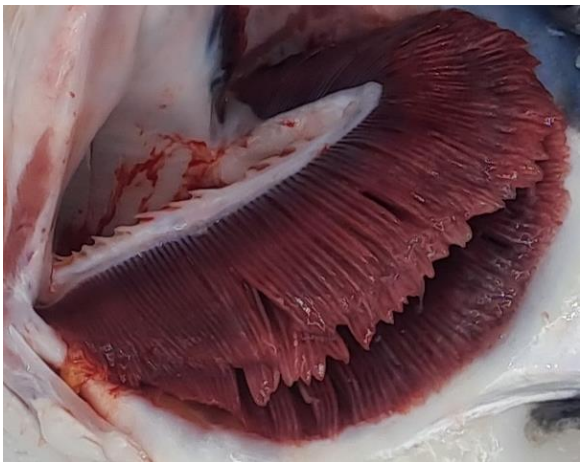
K. Day 23 ICEC, ice storage

L. Day 23 SUBC, ice storage

Figure 10. Variation in eye pupil quality parameter during the ice storage period for both groups of salmon.

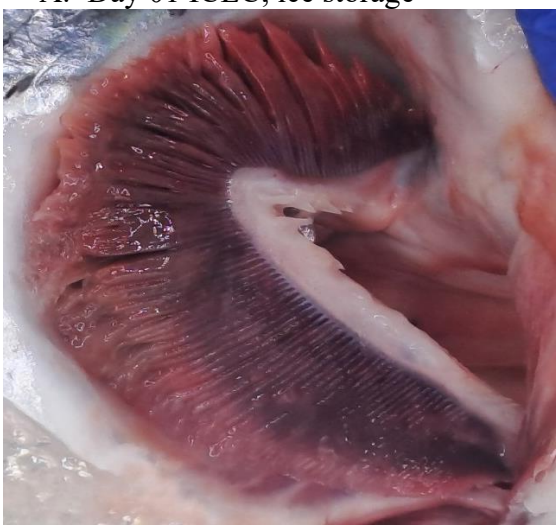
The average scores for the eye pupil colour quality parameter showed a clear linear relationship with the days in ice storage for both the ICEC and SUBC groups of salmon, with R-squared values of R^2=0.856 and R^2=0.912, respectively. Throughout the experimental days, the SUBC salmon group consistently had higher average scores for eye pupils, except on day 16, when the average scores for both groups were almost the same. By the end of the experiment on day 23, the average score for eye pupils was comparatively lower than the average score on day 21 for both ICEC and SUBC salmon. The eye pupil scoring regime varied from 0 to 2, where a score of 0 was given when the eye pupils appeared clear and black with a metallic shine, a score of 1 was assigned if the eye pupils turned dark grey, and a maximum score of 2 was given if the eye pupils turned to a matte grey appearance. From days 1 to 14 in ice storage, the average scores for eye pupils gradually increased, with the SUBC salmon group consistently showing higher average scores than the ICEC group. On day 16, the average score for eye pupils was almost the same (0.7) for both groups. Subsequently, the average scores for eye pupils gradually increased on days 18 and 21 for both ICEC and SUBC salmon. The maximum average score for eye pupils, reaching 1.6, was recorded on day 21 in the SUBC group (Figure 7 E, Table 7).

The eye form, a QI quality factor in the QIM scheme, also exhibited a clear linear relationship with the storage period. However, the average eye form scores fluctuated during the ice storage period for both the ICEC and SUBC groups. The R-squared values for both the ICEC and SUBC groups were R^2 = 0.867 and R^2 = 0.886, respectively. Zero scores for eye form were allocated for a convex appearance as very fresh, while a score of 1 was given when the eye cornea became flat. A maximum score of 2 was assigned to the eye form when it appeared sunken. On day 1 of ice storage, the average eye score was 0.3 and 0.4 for the ICEC and SUBC salmon groups, respectively. By day 4, the average score for eye form increased to 0.7 to 0.9 in both the ICEC and SUBC groups. On day 8 of ice storage, the average score for eye form was almost the same for both ICEC and SUBC salmon. However, on days 11 and 14, the average eye form score was higher in the SUBC salmon group than in the ICEC salmon group. Conversely, on day 16, the results were reversed. The maximum average score of 1.6 for eye form was recorded for the SUBC group on day 18 of storage. However, on days 21 and 23, the average scores for eye form were almost the same for both the ICEC and SUBC groups (see Figure 7 E, Table 7).

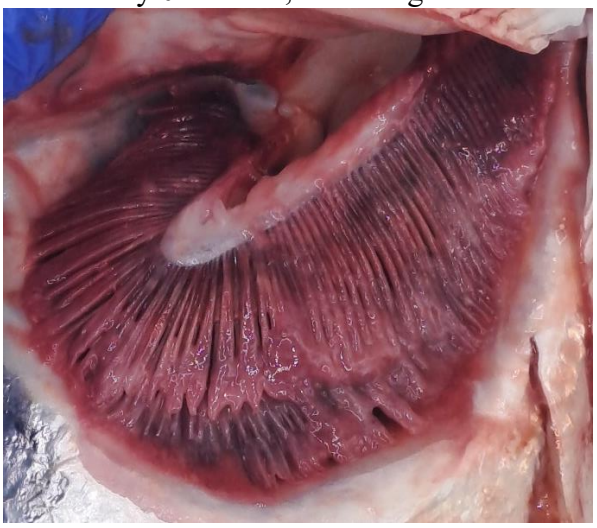


A. Day 01 ICEC, ice storage

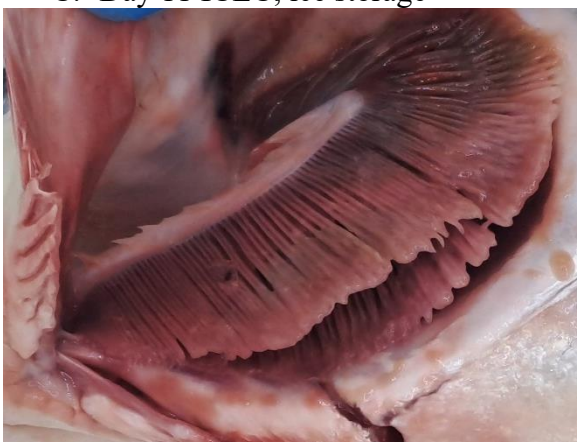
B. Day 01 SUBC, ice storage



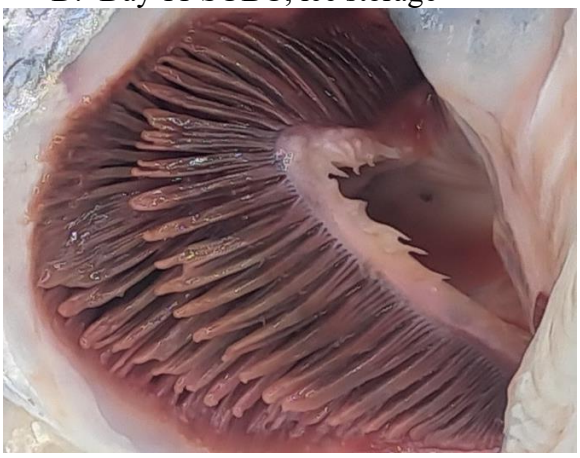
C. Day 11 ICEC, ice storage



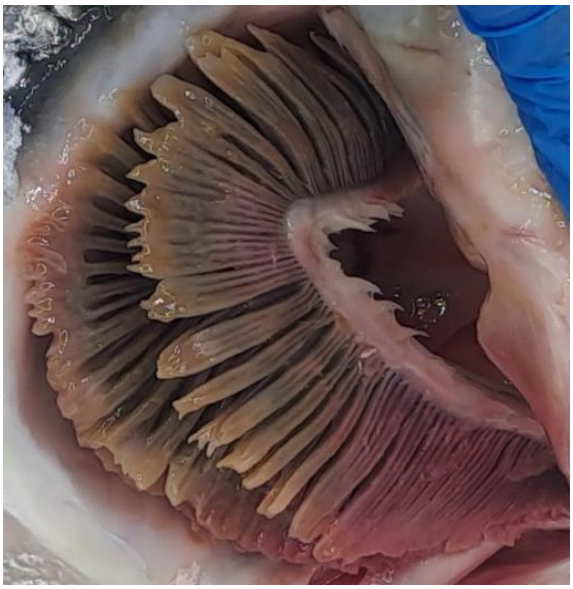
D. Day 11 SUBC, ice storage



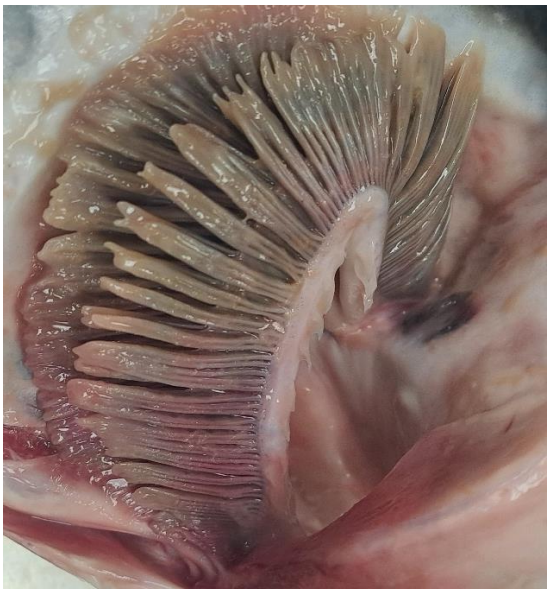
E. Day 14 ICEC, ice storage



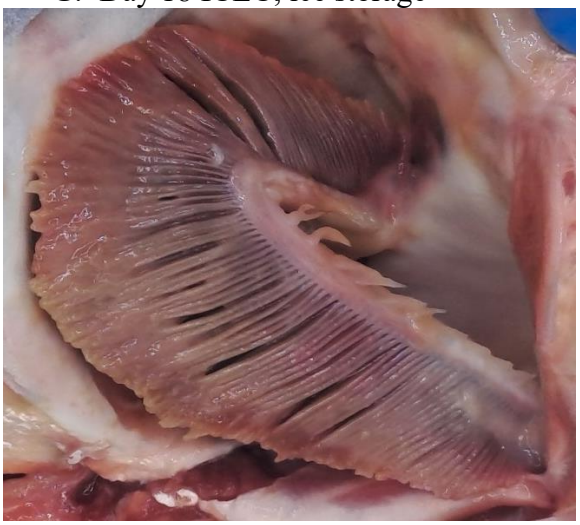
F. Day 14 SUBC, ice storage



G. Day 18 ICEC, ice storage

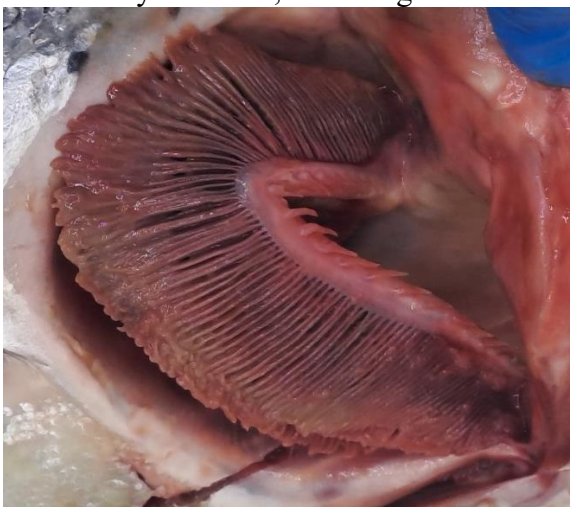


H. Day 18 SUBC, ice storage

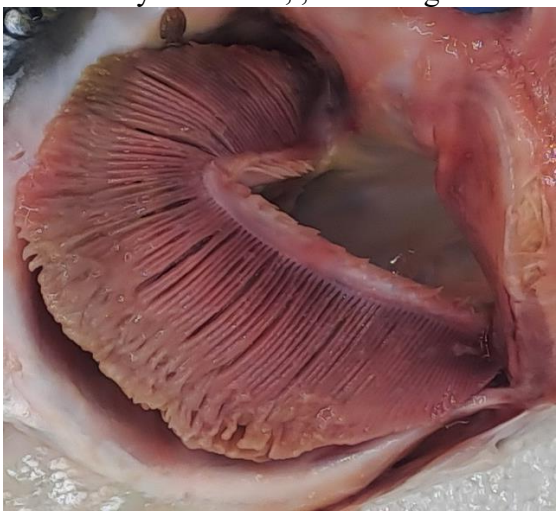


I. Day 21 ICEC, ice storage

J. Day 21 SUBC, , ice storage



K. Day 23 ICEC, ice storage



L. Day 23 SUBC, ice storage

Figure 11. Gill colour/appearance quality parameter variation during the ice storage period for both groups of salmon.

The average scores for the gill colour attribute also showed a linear relationship with days in ice storage. The R-squared values for both the ICEC and SUBC salmon groups were R^2=0.918 and R^2=0.782, respectively. However, the gill colour attribute of the ICEC group indicated a stronger linear relationship than that of the SUBC group during the days of ice storage. The gill colour/appearance scoring regime varied from 0 to 2 as follows: a score of 0 was allocated to the very fresh stage of gill colour, which is red or dark brown in appearance; a score of 1 was given when the gill colour turned to pale red, pink, or light brown; and a score of 2 was allocated if the gill colour turned to grey-brown, brown, grey, or green. On day 1, the average score for gill colour was almost the same in both the ICEC and SUBC salmon groups in ice storage. By day 8, the average score for gill colour doubled (1.6) compared to the average score on day 11 (0.8). On day 16, the average gill colour score was almost the same for both the ICEC and SUBC groups of salmon stored in ice. Subsequently, on days 18, 21, and 23, the average gill colour scores were 1.6, 1.8, and 1.6, respectively, in the ICEC salmon group. The maximum average score for gill colour (1.8) was recorded for the ICEC salmon group on day 21 of ice storage. The average gill colour scores for the SUBC salmon groups on days 18, 21, and 23 of ice storage were 1.3, 1.5, and 1.5, respectively. By the end of day 23, the average gill colour score was almost the same for both the ICEC and SUBC salmon groups in ice storage.

The average scores of the gill mucus attributes for both ICEC and SUBC salmon groups also exhibited a clear linear relationship with days in ice storage. The R-squared values were 0.837 and 0.908 for the ICEC and SUBC salmon groups, respectively. According to the QIM scheme, the gill mucus score regime varied from 0 to 2, with a score of 0 given to transparent gill mucus, a score of 1 given when the gill mucus appeared milky and clotted, and a maximum score of 2 given when the gill mucus colour changed to brown and clotted. During the experimental period, on days 1, 4, and 8 in ice storage, the average scores for the gill mucus attribute were almost the same for both ICEC and SUBC salmon groups, gradually increasing with time in ice storage. On day 11, the average score for gill mucus increased to 1.4 in the ICEC group and was 1.0 in the SUBC group. On day 14, the average score for gill mucus was almost the same for both ICEC and SUBC salmon groups in ice storage. On days 16, 18, 21, and 23, the average scores for gill mucus were almost the same for both ICEC and SUBC salmon groups, gradually increasing with time. The maximum average score for gill mucus was recorded for the SUBC group on day 23 of ice storage.

The average scores for the gill odour quality parameter in both ICEC and SUBC salmon groups showed a better linear relationship with days in ice storage, yielding R-squared values of 0.963 and 0.979 for the ICEC and SUBC salmon groups, respectively. In the QIM scheme, gill odour attribute scores were allocated from 0 to 3, where a score of 0 was given for a fresh seaweed odour from the gills, a score of 1 was assigned when the gill odour turned to a metallic or cucumber odour, a score of 2 was allocated when the gill odour turned to a sour or mouldy smell, and a maximum score of 3 was assigned for a rotten odour from the gills. On days 1 and 4 of ice storage, the average score for gill odour was almost the same in both ICEC and SUBC salmon groups and was very close to 0. However, on days 8, 11, 14, 16, 18, 21, and 23 of the ice storage period, the average score for gill odour was consistently higher in the ICEC group than in the SUBC group. Nevertheless, on day 18 of ice storage, the average score for gill odour was very close between both groups of salmon. The maximum average score for gill odour was recorded for the ICEC salmon group on day 23 of ice storage, representing a value of 2.3.

The 10th attribute in the QIM scheme, the blood in abdomen parameter, also showed a linear relationship with days in ice storage, though with relatively low R-squared values of 0.705 and 0.770 for the ICEC and SUBC salmon groups, respectively. The blood abdomen attribute score

regime ranges from 0 to 1, with a score of 0 given when the blood is red or not present in the abdominal cavity, and a score of 1 when the blood colour changes to brown or yellowish. The average scores for the blood in the abdomen feature varied between the ICEC and SUBC groups. On days 1, 4, and 8, the average score for blood in the abdomen was almost the same in both the ICEC and SUBC groups. However, on day 11, the average score for abdominal blood in the SUBC salmon group increased compared to that in the ICEC salmon group. On days 16, 18, and 21, the average score for abdominal blood was higher in the SUBC group than in the ICEC group. However, on day 23, the average score of 0.6 for blood in the abdomen in the ICEC salmon group was the highest.

The abdomen odour, the 11th QI parameter in the QIM scheme, also exhibited a clear linear relationship with days in ice storage, with R-squared values of 0.939 and 0.952 for both the ICEC and SUBC salmon groups, respectively. The abdominal odour score regime varied from 0 to 3, where a score of 0 was given when the abdominal odour was neutral, a score of 1 was allocated when the abdominal odour turned to cucumber or melon scent, a score of 2 was assigned for a sour odour reminiscent of fermentation, and a maximum score of 3 was given if the odour turned to a rotten or rotten cabbage scent in the abdominal cavity. However, during the experimental period, on most days of ice storage, the average abdominal odour score was almost the same for both the ICEC and SUBC salmon groups, except on days 11 and 16. The maximum average score for abdomen odour (1.7) was recorded for the ICEC salmon group on day 23 of storage in ice.

4.2.2 GDA

The GDA odour and flavour sensory attribute scores and p-values for both ICEC and SUBC cooked salmon are presented in Tables 8 and 9, respectively.

Table 8. Odour sensory attribute scores and p values for SUBC and ICEC cooked salmon.

Date	**Days in ice storage**	**Group**	**Odour characteristics**						
			Fresh			**Spoilage**			
			Sweet	**Oil**	**Sour**	**Rancid**	**Queasy**	**Putrid**	**Spoilage**
05.03.2024	*day 1*	ICEC	53	27	0	1	0	0	0
		SUBC	44	27	0	0	0	0	0
		p-value	0.093	0.937	0.356	0.216	-	-	-
15.03.2024	*day 11*	ICEC	50	17	1	5	4	0	4
		SUBC	42	17	1	5	3	0	5
		p-value	0.139	0.911	0.394	1.000	0.423	-	0.647
20.03.2024	*day 16*	ICEC	50	16	0	1	1	0	1
		SUBC	45	17	0	2	1	0	1
		p-value	0.048	0.544	-	0.325	0.351	-	0.227
22.03.2024	*day 18*	ICEC	44	12	1	2	2	0	3
		SUBC	46	15	0	5	0	0	2
		p-value	0.644	0.131	0.363	0.363	0.363	-	0.505
25.03.2024	*day 21*	ICEC	34	11	2	4	3	0	5
		SUBC	44	16	2	2	4	1	5
		p-value	0.008	0.093	0.542	0.315	0.730	0.363	0.890
27.03.2024	*day 23*	ICEC	40	22	1	4	2	1	4
		SUBC	36	17	2	4	1	1	4
		p-value	0.426	0.394	0.399	0.604	0.391	0.391	0.807

Table 9. Flavour sensory attribute scores and p-values for SUBC and ICEC cooked salmon.

Date	Days in ice storage	Group	Flavour characteristics					
			Fresh		Spoilage			
			Sweet	Oil	Sour	Rancid	Queasy	Spoilage
05.03.2024	*day 1*	ICEC	50	29	1	0	0	0
		SUBC	51	31	1	0	0	0
		p-value	0.855	0.675	1.000	-	-	-
15.03.2024	*day 11*	ICEC	36	23	0	2	2	2
		SUBC	41	26	1	1	2	1
		p-value	0.431	0.565	0.304	0.624	0.363	0.745
20.03.2024	*day 16*	ICEC	47	19	0	0	1	0
		SUBC	43	16	0	1	1	1
		p-value	0.095	0.135	-	0.125	0.944	0.061
22.03.2024	*day 18*	ICEC	40	15	1	0	2	2
		SUBC	46	17	1	3	0	1
		p-value	0.204	0.376	1.000	0.363	0.363	0.482
25.03.2024	*day 21*	ICEC	39	15	1	2	2	2
		SUBC	47	17	2	2	2	3
		p-value	0.058	0.487	0.287	1.000	0.472	0.536
27.03.2024	*day 23*	ICEC	37	23	1	1	3	2
		SUBC	41	21	2	1	2	2
		p-value	0.033	0.663	0.614	0.391	0.182	0.861

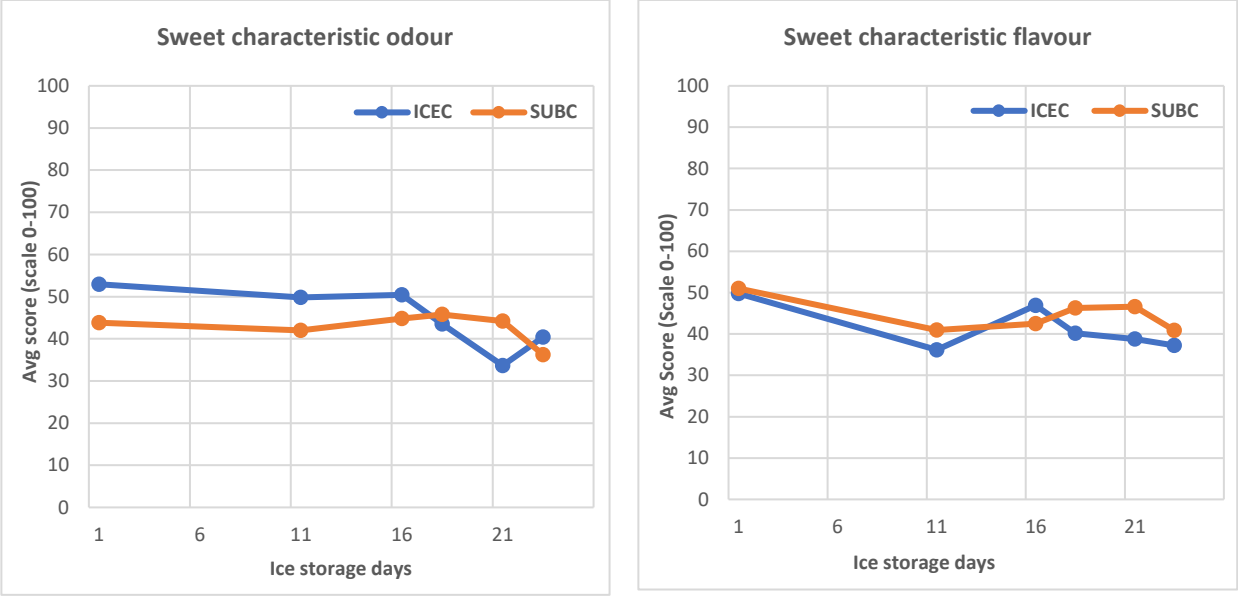
The sensory panel employed Generic Descriptive Analysis (GDA) to evaluate the parameters of odour and flavour. According to the results obtained from the GDA, freshness attributes remained dominant throughout the storage period for both ICEC and SUBC salmon groups. The most dominant attribute was the sweet characteristic of the odour and flavour for both groups. The average scores for the freshness attribute of sweet odour and flavour ranged from 36 to 53 and 36 to 51 for the ICEC and SUBC groups, respectively, throughout the ice storage period (Tables 8 and 9). On storage day 21, the sweet flavour was close to being significantly higher for the SUBC group, and on storage day 23, the sweet flavour was significantly higher in the SUBC group (Figure 9). By the end of storage day 23, the second freshness attribute of fresh fish oil odour and flavour was observed, with values that were comparatively lower than the sweet characteristic attributes. The average scores for the freshness attribute of fresh fish oil odour and flavour ranged from 11 to 27 and 15 to 31 for the ICEC and SUBC groups, respectively, throughout the ice storage period (Table 08 & 09).

However, the characteristic salmon and oily flavours were clearly mentioned by the panellists, indicating a difference between the groups. At the end of the evaluation of the cooked salmon, a rejection point was not reached, indicating that the end of the shelf life was not obtained. Odour and flavour attributes with freshness characteristics of fish oil were detected at a constant level with slight fluctuations throughout the days in cold storage in ice for both groups of ICEC and SUBC salmon. The sweet attribute remained dominant throughout the storage period (Tables 8 and 9).

Attributes such as sour, rancid, queasy, and putrid, which are negative and indicate spoilage characteristics, were detected at very low levels or were undetected throughout the storage period, with GDA scores below 10. The average scores for the freshness attribute decreased

slightly over time for both groups of salmon. However, at the end of the evaluation of cultured Atlantic salmon in both the ICEC and SUBC groups, the end of shelf life was not reached.

Figure 12 illustrates the mean values for GDA sensory attributes of the most dominant attributes sweet odour and flavour for cooked Atlantic salmon in ICEC and SUBC groups.



(A) Sweet characteristic odour (B) Sweet characteristic flavour

Figure 12: Freshness characteristic odour (A) and flavour (B) scores for cooked ICEC and SUBC salmon groups during ice storage.

On storage day 23, three salmon from the ICEC group and three from the SUBC were filleted. The odour of three raw fillets from each group was evaluated by two trained assessors. Fillet examination was conducted to better understand the condition of the flesh, especially in the tail area of the fish. The results are presented in Table 10.

Table 10. Description of the sensed odour attributes of salmon fillets.

#	Group	Code	Description for odour attribute from fillet
1	ICEC	187	Sour odour from entire fillet
2		227	Cucumber odour from middle portion and sour from tail region
3		379	Cucumber odour from middle portion and sour from tail region
4	SUBC	447	Cucumber odour from entire fillet
5		597	Cucumber odour from middle portion and sour from tail region
6		671	Cucumber odour from entire fillet

All three fillets in the ICEC group had a sour odour from the tail region, and in one of the fillets, a sour odour was detected from the entire fillet, whereas a cucumber odour was detected from the middle portion of the other two fillets. Additionally, a sour odour was detected in the tail region of two fish (227 and 379) in the ICEC salmon group. The fillets in the SUBC group all had cucumber odour, although a sour odour was detected in the tail part (Table 10).

4.3 Microbial analysis

Microbial counts in both the ICEC and SUBC groups increased with storage time (Figure 13). At the initial measurement point on storage day 1, the Total Viable Count (TVC) for both groups of ICEC and SUBC was less than 10 CFU/g (log CFU/g= 1.0). However, after 21 d of storage on ice, the TVC values for ICEC salmon exceeded 1.1 × 10^5 CFU/g (log CFU/g= 5.0) and 1.9 × 10^5 CFU/g (log CFU/g= 5.3) on day 23 (end of storage). The TVC value for the SUBC group was approximately 2.6 × 10^4 CFU/g (log CFU/g= 4.4) on day 23.

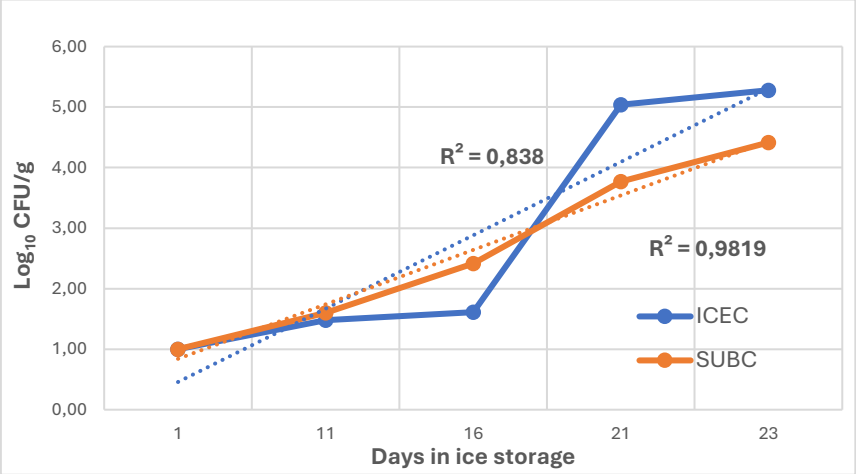


Figure 13. Variation in TVC counts with days of ice storage for ICEC and SUBC salmon.

A higher correlation (R^2 = 0.9819) was observed between the days in ice storage and log total viable counts for the SUBC salmon group during ice storage, and a weaker correlation (R^2 = 0.838) was observed for the ICEC salmon group in ice storage (Figure 13).

5 DISCUSSION

This study focused on whether there was a need to adjust the existing Quality Index Method (QIM) scheme developed for farmed Atlantic salmon stored in ice to more accurately evaluate the freshness of sub-chilled salmon. This was done by monitoring the changes in sensory attributes of whole gutted salmon with the QIM scheme, changes in sensory attributes of cooked salmon with Generic Descriptive Analysis (GDA), and microbial quality by Total Viable Counts (TVC) during storage for up to 23 days. For this purpose, subchilled (SUBC) and conventional ice-chilled (ICEC) gutted salmon were stored in ice in Styrofoam boxes. The SUBC group was kept in cold storage at -2°C to 0°C for up to 16 days, when it was transferred to cold storage at0°C to 4°C until the end of the storage study of 23 days. This was done because refrigerated cool rooms are typically used for shipping freight for 14-day period. The ICEC group was kept in cold storage at 0°C to 4°C from storage day 1 until 23 days. The temperature logger placed outside the boxes of ICEC in the cool room was erased, but the logger placed outside the boxes for the SUBC data was recovered. The temperature range was -2.5 to -0.1 °C, and the average temperature was - 0.8°C ± 0. 8.

The average temperature recorded for the core of salmon fish was - 0.9°C ± 0.1 for SUBC salmon and 0.4°C ± 0.2 for ICEC salmon during the ice storage period. The temperature range (core of fish) also varied, with SUBC salmon ranging from - 1.0°C to -0.4°C and ICEC salmon ranging from 0. 4°C to 0.5°C. According to the Arnarlax report provided with the salmon samples, the indicated average core temperature for SUBC salmon was -0.4°C ± 0.0, and for ICEC salmon, it was 1.9°C ± 0.1. However, during the experimental period, the core temperature logger readings showed an average of 0.5°C higher for SUBC salmon and 1.5°C lower for ICEC salmon than the reported values. This indicates that the temperature of the core of fish for both groups of SUBC and ICEC salmon during the ice storage period was well-controlled in a successful manner.

The assessment of individual panellist performance during the sensory evaluation with QIM demonstrated some variation between assessors. Some assessors scored the samples consistently higher or lower throughout the shelf-life experiment. Earlier trials have shown similar results to those of Sveindottir *et al*. (2002) and Sveindottir *et al*. (2003).

Throughout the 23-day experimental period, the overall average scores for the quality parameters of skin odour, skin texture, eye form, gill mucus, blood in the abdomen, and abdomen odour were nearly identical in both ICEC and SUBC salmon groups. However, there was a consistent trend of higher average scores for the eye pupil attribute within the QIM scheme for SUBC than for ICEC throughout the experiment. After a storage period of 16 d on ice, the average skin mucus and gill colour scores in the ICEC group were comparatively higher than those in the SUBC group. Further analysis revealed a clear linear relationship between the average QI values of both the ICEC and SUBC groups of salmon and the duration of ice storage, as assumed. However, no statistically significant difference was observed between the ICEC and SUBC Atlantic salmon groups across various ice storage dates. Nevertheless, on days 01, 04, and 08, which marked the initial stages of ice storage, the average QI values for the SUBC group were higher than those of the ICEC group. By day 11, the average QI values were similar in both groups. Subsequently, from days 16 to 23 (the end of the experiment), the average QI values of the ICEC group were consistently higher than those of the SUBC group. In general, during the initial days, approximately 14 days, the SUBC group exhibited higher QI values than the ICEC group. However, as the storage duration progressed, particularly after 16-day mark, the average QI values for the ICEC group surpassed those of the SUBC group.

The microbiological analysis conducted in this experiment yielded results similar to the Total Viable Counts (TVC) of salmon flesh observed in the study by Sveinsdottir *et al*. (2002), where the point of rejection was reached at 20 days for ICE salmon. In comparison, the ICEC salmon was microbiologically rejected when the count exceeded the 10^5 CFU/g (log CFU/g = 5.0) threshold in the flesh of whole salmon, occurring at the point of rejection at 20 days (Sveinsdottir *et al*., 2002). However, in this study, the endpoint of rejection was not reached for SUBC. Remarkably, ICEC salmon exceeded its shelf life, lasting up to 23 days under ice storage conditions. Based on microbial aspects, the shelf life of SUBC salmon could potentially be longer than 23 d under similar ice storage conditions.

In the GDA analysis, the freshness attributes of odour and flavour were consistently detected throughout the experimental period, albeit decreasing over time. Conversely, attributes such as sour, rancid, queasy, and putrid, indicative of spoilage, were either detected at very low levels or were undetected throughout the storage period. At the end of the experiment, neither the ICEC nor the SUBC group of cultured Atlantic salmon reached the end of shelf life on storage day 23. According to Sveindóttir *et al*. (2002 and 2003), in QDA analysis, positive attributes for freshness, odour, and flavour were obtained at the beginning of ice storage. However, after 21-22 days of ice storage, they were hardly detectable, and the salmon reached marginal acceptability as negative attributes dominated.

Carotenoid red pigments (Sveindottir *et al*., 2002), such as astaxanthin (Sebra & Pedrosa, 2010), naturally occur in salmon flesh and are believed to play a crucial role in protecting lipid tissues by inhibiting lipid oxidation (Sveindottir *et al*., 2002; Sebra & Pedrosa, 2010). Both studies cited this as the reason for the low rancidity observed during the 21-day ice storage period. The feed given to farmed salmon often includes carotenoids to enhance this quality (Sveindottir *et al*., 2002). Additionally, Sveinsdottir *et al*. (2002) identified propionaldehyde and (Z)-1,5-octadien-3-one as the most potent high-volatile odourants in cooked fresh salmon samples. These compounds imparted a sweet and metallic odour, contributing to the overall flavour profile of the cooked salmon. Furthermore, according to Sveinsdottir *et al*. (2002), various odourants from fresh cooked salmon were described, with the characteristic salmon odour attributed to compounds such as propionaldehyde and acetaldehyde for sweetness, hexanal and (Z)-3-hexenal for green leaves smell, methional for boiled potato-like aroma, dimethyl trisulfide for cabbage-like scent, and 1-octen-3-one for mushroom-like odours. The oily flavour and aroma might have been attributed to (Z, Z)-3,6-nonadienal, which is described as fatty and green (Sveinsdottir *et al*., 2002).

To better understand the situation, it is important to identify both internal and external changes that may occur in the salmon's body and the areas where factors are involved in inducing these changes. Numerous studies have explored the biological, chemical, and physical factors affecting the cold storage shelf-life of salmon (Doyle (1989); Sveinsdottir *et al*. (2002); Martinsdottir *et al*. (2003); Bahuaud *et al*. (2008); Duun and Rustad (2008); Sebra and Pedrosa (2010); Kaale (2011 and 2013); Gökoğlu and Yerlikaya (2015); Kaale and Eikevik (2016); Ochiai and Ozawa (2020)) and other species (Chang *et al*. (1998); Nielsen *et al*. (2004); Lushchark (2007); Devhastin *et al*. (2010); Provincial *et al*. (2010); Dowlati *et al*. (2013); Osibona and Ezekiel (2014)). Biological factors associated with salmon fish encompass various aspects, including the size, sex, maturity stage, individual biological features, parasitic attachment, fish tissue anatomy and structure, rigor stage, muscle thickness, and lag phase of spoilage microbes. These factors collectively contribute to the overall quality and shelf life of salmon. Chemical factors also play a crucial role and are influenced by factors such as the fish feeding regime during aquaculture, nutritional composition of the fish, protein content, fat

content, water content, oxidation states, protein denaturation, and pH in muscle tissue. These chemical parameters can significantly affect the quality and preservation of salmon. In addition, physical factors such as temperature, cell tissue fluid, and drip loss are noteworthy contributors. For instance, temperature fluctuations can affect enzymatic and microbial activity, thereby influencing the rate of spoilage. Variations in cell tissue fluid and drip loss can impact the texture and overall freshness of salmon.

Considering the comprehensive interplay of these biological, chemical, and physical factors, it is evident that one or more of these factors, either individually or in combination, may affect salmon quality, even under optimal preservation conditions. Understanding the complex dynamics among these factors is essential for enhancing the shelf life and maintaining the quality of salmon during its storage. These factors directly or indirectly influenced the quality of both the ICEC and SUBC salmon groups, as well as the quality parameters measured using sensory and microbiological evaluations. Among these factors, temperature plays a pivotal role in cold preservation techniques, such as chilling, superchilling, and freezing.

At super-chilling temperatures, microbial activity is reduced, and most bacteria cannot grow (Kaale *et al*., 2011). However, autolytic, enzymatic, or other chemical reactions may occur at a higher rate or even accelerate (Magnussen *et al*., 2008). Consequently, initially, both the ICE and SUB group salmon showed low Total Viable Counts (TVC) values (log CFU/g), but in the later days (21 and 23), the TVC value in ICEC salmon increased and reached rejection levels. Kaale and Eikevik (2015) noted that at traditional chilling temperatures, many spoilage microorganisms remain highly active and pathogenic bacteria can grow and produce toxins. Therefore, lowering the temperature as much as possible without damaging the product is crucial. Superchilling, which combines partial freezing and storage at low temperatures, extends the shelf life of products by drastically reducing microbial activity (Duun and Rustad, 2008).

Additionally, in the initial stages, microbial growth may be in the lag phase due to temperature inhibition of bacterial activity, particularly their biological functions. Doyle (1989) emphasised that the shelf life of seafood is primarily determined by temperature, which controls the rate of bacterial spoilage, enzymatic activity, and fat oxidation. The TVC of SUBC salmon did not reach the rejection limit during the 23-day ice storage period, gradually increasing with time, indicating that the shelf life of SUBC salmon would likely be reached after a few more days.

Temperature is intricately linked with other factors and can influence their activities, either accelerating or inhibiting them, thereby affecting the quality index (QI) parameters and the attributes evaluated through GDA. According to Duun (2008), super-chilling extends the shelf life of food compared to conventional chilling and is superior to freezing, as a larger proportion of the bulk water remains unfrozen compared with frozen products. This results in less structural damage, a lower degree of freeze denaturation, and reduced drippage loss. Similarly, Kaale and Eikevik (2015) observed numerous advantages of super-chilled foods over traditional chilling or freezing methods. These findings underscore the significance of temperature control, particularly in superchilling processes, for preserving the quality and extending the shelf life of food products.

The Quality Index Method (QIM) scheme developed for cultured Atlantic salmon is primarily tailored for assessing fish stored in ice, with quality parameters, especially appearance, relying heavily on visual assessments. Newly caught fish typically exhibit a clearer appearance and receive lower scores. However, when stored in ice or ice-chilled environments, the natural appearance of the fish is generally preserved because the temperatures hover around or slightly

below the initial freezing point (-0.5 to -2.8°C for most foods). In the super-chilling process, the temperature of the food product is lowered to 1-2°C below this range (Duun and Rustad, 2007). Therefore, the appearance of SUBC salmon may be affected. According to Kaale (2013), this initial surface freezing results in an approximately 2 mm layer of ice along the product surface. Magnussen *et al*. (2008) also mentioned a 1–3 mm ice layer formed after super chilling. While this may be advantageous in achieving temperature equilibrium between the exterior and interior during storage, it can alter the external appearance of salmon. As a result of the formation of a 2 mm or thicker ice layer, some appearance quality parameters may exhibit higher scores for SUBC than for ICEC salmon. Consequently, the Quality Index (QI) values for SUBC salmon were higher than those for ICEC salmon, reflecting the differences in appearance caused by the super-chilling process. This could explain the higher values observed for SUBC salmon in the initial days of storage compared to ICEC salmon.

Colour is one of the most crucial quality parameters, as it is one of the first things consumers notice. Salmon skin contains black spots formed by specialised chromatophores called melanophores (or melanocytes), which produce dark pigments known as eumelanin (Bagnara and Matsumoto, 2006). These pigments, along with the proteins they produce, can be damaged or become inactive when suddenly exposed to the super-chilling process. Pigment denaturation may also occur. However, when subjected to ice chilling, the pigments were less likely to be harmed. Therefore, differences in skin appearance may have occurred in the SUBC salmon group due to the super-chilling process. In later days, light grey patches were observed on the skin, possibly due to pigment denaturation or skin deterioration. Over time, the appearance may decrease, resulting in higher Quality Index (QI) scores for both groups of salmon. Furthermore, other factors such as exposure to stress conditions in aquaculture tanks while alive, slaughtering time and method, individual biological features, and rigor stage can also affect skin appearance of the fish. Additionally, the feeding regime and composition of feeds during the fish's lifetime could influence skin colouration. These factors collectively contribute to the overall appearance and quality of salmon.

The appearance quality parameter of gill colour in the QIM scheme exhibited variations between the two groups. The individual scores for gill colour significantly impacted the ultimate QI value in both groups. Gill colour also varied over time, with SUBC salmon initially receiving higher scores than ICEC salmon, and vice versa in later days. The gills of fish serve as the main respiratory organs and are regularly exposed to the external environment. Their structure consists of thinner tissue with a high blood supply, making them susceptible to spoilage owing to biological, chemical, and physical factors. The gills are composed of filamentous proteins with two or three cell layers for osmoregulation with blood and water. Super-chilling conditions can directly affect gill tissues by denaturing proteins, causing structural damage, and rupturing thin tissues. Initially, internal damage may not be visibly apparent, resulting in lower gill appearance scores. However, as tissue damage progresses, microbial activity and chemical reactions occur, leading to higher gill quality scores. These observations highlight the complex interplay between superchilling conditions, gill tissue integrity, and overall salmon quality.

Eye pupils and eye shape are also significant quality parameters primarily considered by consumers. It is likely that the SUBC salmon group would receive a higher score when assessed using QIM. As mentioned earlier, the denaturation of proteins and other structural compounds can lead to a loss of quality compared to that of ICEC salmon. The eye is a thin and sensitive tissue structure, similar to gill tissues, and is susceptible to damage. The formation of an ice crystal layer on the body can disrupt its visual appearance. Consequently, changes in the colour of the eye pupils may occur, and the cornea of the eye can easily become contracted and sunken

when subjected to lower temperatures during super chilling. These factors contributed to the variations in eye appearance between the ICEC and SUBC salmon groups, further emphasising the impact of preservation methods on the overall quality of salmon.

The odour and flavour attributes of the flesh of both ICEC and SUBC salmon fell within the freshness range and were observed throughout the experimental period of 23 days. Neither salmon group was rejected during the GDA analysis, confirming an extension of shelf life for the salmon flesh of more than 23 d. Several factors may have contributed to the increase in shelf life. Chemical compounds that naturally occur in salmon and are generated after the fish's death due to enzymatic and chemical reactions, such as lipid oxidation and breakdown into primary and secondary products, as well as microbial secretions, may influence the taste and smell of the fish. Astaxanthin, a carotenoid red pigment naturally found in salmon flesh, is one such compound that can affect the shelf life of salmon. Astaxanthin possesses significant antioxidant properties that help mitigate the detrimental effects of oxidation in food products. It is typically acquired by salmon through their diet, originating from microalgae, zooplankton, insects, or crustaceans.

As mentioned earlier, colour is the first impression from the customer's perspective, and to enhance the flesh colour of salmon, industries may add astaxanthin or similar carotenoids to artificial salmon feeds. Therefore, feeding regime and feed composition also play a role in affecting the shelf life of salmon, given the competitive nature of the industry. Seabra and Pedrosa (2010) suggested that the addition of astaxanthin could further prolong the shelf life of products.

The flesh of salmon is considered a rich nutritional source, particularly high in protein and fat content. Proteins can undergo various changes, such as oxidation reactions, freeze denaturation, or interactions with lipid oxidation products, all of which tend to render proteins insoluble (Lushchark, 2007). Freezing a product can induce freeze denaturation of proteins due to several stresses, including ice formation, changes in solute concentration resulting from water crystallisation, and subsequent pH alterations (Lushchark, 2007). Moreover, the oxidation of myofibrillar proteins increases during the storage of postmortem muscles (Martinaud *et al*., 1997). Additionally, Bhat *et al*. (2018) explained that oxidation in postmortem muscle also affects protein activity by either inactivating or modifying calpain activity, which is an intracellular enzyme system.

The structure of muscle tissue fractures from ice crystal formation and the breakdown of myofibrillar proteins, causing cell contents to leak out along with extracellular fluid, contributing to drip loss. Many nutrients are water-soluble, exiting fish tissue in drip loss and serving as food for microorganisms, which proliferate faster due to easy access to oxygen. This accelerated microbial growth shortens the shelf life of the product. Moreover, as water freezes, it damages cell structures and increases the solute concentration of the remaining water, resulting in more available water being lost as drip loss during thawing.

Fatty acids, which are crucial components of many lipids, significantly contribute to the properties of food owing to their structure. Lipid oxidation, also known as oxidative rancidity, occurs when oxygen interacts with unsaturated fatty acids in an autooxidation reaction, resulting in the formation of several chemical products associated with product degradation. This reaction is highly undesirable and is prevalent in foods rich in polyunsaturated fatty acids (PUFA) because the α-methylene groups (the CH2-groups adjacent to the double bond) are susceptible to oxidation. During storage, there is a delay before primary oxidation products

accumulate, measured via PV-titration. The rate of primary oxidation buildup is expected to rise after some days of storage, followed by a decline as primary oxidation products transform into secondary ones. Consequently, one can anticipate an initial increase in PV values followed by a decrease as secondary oxidation products accumulate. These factors directly or indirectly affect the quality parameters and GDA attributes.

Many nutrients in the salmon body are water-soluble, and after breakdown, protein-like compounds exit the fish tissue in drip loss, serving as food for microorganisms. This accelerated microbial growth shortens the shelf life of the product. The structure of muscle tissue fractures from ice crystal formation and the breakdown of myofibrillar proteins, causing cell contents to leak out along with extracellular fluid, contributing to drip loss. As water leaks out, various compounds exit with it, including sarcoplasmic, water-soluble proteins (Devahastin, 2010).

The pH and ice crystal formation significantly influence drip loss, as most of the bulk water shelves within the myofibrillar network. Larger ice crystals can break down more muscle tissue, leading to leakage as the network collapses (Devahastin, 2010). A decrease in muscle pH creates an acidic environment, triggering the release of lysosomal cathepsin, which further breaks down muscle tissue. Consequently, the network holding the water collapses, resulting in water release as drip loss. Protein denaturation may occur during superchilling. This could be the reason for the higher QI value for SUBC salmon at later dates of ice storage.

Ultimately, the size, sex, and maturity stage of fish also affect their quality and shelf life. Previous studies conducted by Sveindóttir *et al*. in 2002 and 2003 utilised fish with an average weight of 3-4 kg and a size lower than that in this experiment. In this experiment, fish with an average weight exceeding 4 kg were used, and their flesh was comparatively thicker than that of lower-weight salmon. According to Glover *et al*. (2009), differences in salmon flesh quality parameters are closely linked to fish size. Furthermore, according to Glover *et al*. (2009), farmed and wild-caught salmon have average weights of 4433 g and 2013 g, respectively. The percentage of fillets for farmed salmon was 72.8%, whereas that for wild-caught salmon was 71%. Furthermore, the texture (mechanical testing) of farmed salmon is 843 g, whereas that of wild-caught salmon is 807 g. The dry matter content in farmed salmon was 34%, which was slightly higher than the 33.5% found in wild-caught salmon. This may be attributed to the feeding regime and feed composition provided by the industry. Consequently, microbes involved in spoilage may find it difficult to penetrate the thicker flesh layers, resulting in spoilage mainly occurring in the surface layers. Additionally, the formation of a 2 mm-thin layer of ice crystals may further prevent microbial invasion. However, in the tail region, which is thinner than the middle of the salmon, some spoilage attributes were observed during the fillet examination at the end of the experiment. Therefore, when suggesting further studies, the size, sex, and maturity stages of the fish should be considered. Furthermore, as salmon, especially females, reach maturity, their fat content increases over time due to spawning. High fat content can lead to oxidation and the production of primary and secondary metabolic compounds, affecting the fish shelf life. In this study, fish size appeared to contribute to the increased shelf life of both ICEC and SUBC salmon.

6 CONCLUSION

In this experiment, two groups of gutted farmed Atlantic salmon (*Salmo salar*) were evaluated during storage time up to 23 days: one was ice-chilled and stored in ice at 0–4°C (ICEC), and the other was subchilled and stored in ice at -2°C to 0°C (SUBC) until storage day 16, when it was moved to 0–4°C for up to 23 days of storage. The published Euro-fish Quality Index Method (QIM) scheme for farmed Atlantic salmon was used to evaluate the freshness of the whole salmon. The average Quality Index (QI) scores increased linearly with storage time in ice. Throughout the 23-day experimental period, the overall average scores for the individual quality parameters of skin odour, skin texture, eye form, gill mucus, blood in the abdomen, and abdomen odour did not differ between the ICEC and SUBC groups. However, there was a consistent trend of higher average scores for the eye pupil and eye form attributes within the QIM scheme for SUBC salmon compared to ICEC salmon throughout the experiment. After a storage period of 16 days on ice, the average skin mucus and gill colour scores in the ICEC group were comparatively higher than those in the SUBC group. Therefore, it is recommended to change the current quality index method (QIM) scheme for iced farmed Atlantic salmon to focus on subchilled salmon, with less emphasis on eye pupils and eye form. Individual panellists scored the salmon systematically higher or lower than the majority of the panellists. Although guidance material is provided with the QIM, panel training is important. Some panellists did not receive training in evaluating salmon using QIM before the experiment, which might have affected the results.

Neither the sensory evaluation conducted using the QIM for the whole salmon nor the Generic Descriptive Analysis (GDA) for cooked salmon indicated the end of shelf life within the duration of ice storage of 23 days. No spoilage attributes were detected in the cooked samples according to the GDA, but scores for the attributes that were the most characteristic at the beginning of storage (freshness attributes) decreased with storage time. No statistical differences between the iced (ICEC) and sub-chilled (SUBC) were observed in most sensory attributes of odour and flavour, throughout the experiment. The only exception was the sweet flavour on storage day 23, when the subchilled salmon had a higher sweet flavour. Although it cannot be concluded that the groups differ in shelf life based on the GDA results, the results indicated that the sub-chilled group retained freshness characteristics longer than the iced group. The total viable microbial counts in the SUBC group did not reach unacceptable limits but were comparatively higher in ICEC salmon after 21 and 23 days of storage. At the end of the storage trial, on day 23, the ICEC salmon reached an unacceptable microbial quality. Based on the microbial analysis, it can be concluded that salmon stored in ice reached the end of shelf life after 23 days.

Essentially, the SUBC-treated farmed Atlantic salmon exhibited certain advantages over ICEC salmon when evaluated using sensory methods (QIM and GDA) and microbiological analysis. The study indicated that the shelf life of farmed gutted Atlantic salmon with subchilling (SUBC) exceeded 23 days in ice storage, whereas the salmon not undergoing subchilling was at the end of shelf life. According to previous studies, the shelf life of iced Atlantic salmon is approximately 20 days. Factors such as fish size, flesh thickness, nutritional composition, rigor stage, lag phase of microbes, and muscle pH affect shelf life. Future studies should consider the aforementioned factors and increase the number of salmon samples per batch to more than three for each sensory session. Quality Index Method (QIM) and microbial analysis should be conducted every 2-3 days, extending the experimental period to up to 25 or 30 days to enhance precision.

ACKNOWLEDGEMENTS

I would like to thank my supervisors Kolbrún Sveinsdóttir and Aðalheiður Ólafsdóttir for their exceptional advice and guidance rendered to me during my study. Special thanks to Arnarlax for providing the salmon samples free of charge to complete this study. I wish to recognise the commitment of the sensory panel at MATIS, who were an integral part of my experiment. Many thanks to the staff of GRÓ FTP, Thor Heiðar Ásgeirsson, Mary Francis Davidson, Julie Ingham, Stefán Úlfarsson, Davíð Tómas Davíðsson, and Zaw Myo Win, for their periodic support in helping me complete this research and fellowship programme. I am also grateful to the fellows who have, in one way or another, assisted me during this project. Finally, I wish to express my sincere appreciation to the institutions involved: National Aquatic Resources Research and Development Agency (NARA), Sri Lanka, Matis, Iceland, Arnarlax, Iceland, and GRO-FTP, as well as the Government of Iceland, who made my participation in GRÓ FTP 2022 and 2023 possible.

BIBLIOGRAPHY

Ando, M., H. Nakamura, R. Harada, and A. Yamane. 2004. Effect of super-chilling storage on maintenance of freshness of kuruma prawn. Food Sci Technol Res. 10 (1):25–31. doi:10.3136/fstr.10.25.

Ariyawansa, K. W. S., Wijendra, D. N., & Senadeera, S. P. S. D. (2003). Quality Index Method developed for frigate tuna (*Auxis thazard*). *Sri Lanka Journal of Aquatic Sciences, 8*(1), 95–109. https://doi.org/10.4038/sljas.v8i1.7495

Bagnara, J. T., & Matsumoto, J. (2006). Comparative anatomy and physiology of pigment cells in nonmammalian tissues. In J. J. Nordlund, R. E. Boissy, V. J. Hearing, R. A. King, W. S. Oetting, & J.-P. Ortonne (Eds.), *The pigmentary system: Physiology and pathophysiology* (2nd ed.). Blackwell Publishing.

Bahuaud, D., Mørkøre, T., Langsrud, Ø., Sinnes, K., Veiseth, E., Ofstad, R., & Thomassen, M. S. (2008). Effects of −1.5°C super-chilling on quality of Atlantic salmon (*Salmo salar*) pre-rigor fillets: Cathepsin activity, muscle histology, texture and liquid leakage. *Food Chemistry, 111*(2), 329–339. https://doi.org/10.1016/j.foodchem.2008.03.075

Banerjee, R., & Maheswarappa, N. B. (2019). Super-chilling of muscle foods: Potential alternative for chilling and freezing. *Critical Reviews in Food Science and Nutrition, 59*(8), 1256–1263. https://doi.org/10.1080/10408398.2017.1406335

Banerjee, R., & Maheswarappa, N. B. (2017). Super-chilling of muscle foods: Potential alternative for chilling and freezing. *Critical Reviews in Food Science and Nutrition*. https://doi.org/10.1080/10408398.2017.1401975

Bataringaya, A. (2007). *Analysis of quality deterioration at critical steps/points in fish handling in Uganda and Iceland and suggestions for improvement* (Final project). United Nations University Fisheries Training Programme.

Beaufort, A., M. Cardinal, A. Le-Bail, and G. Midelet-Bourdin. 2009. The effects of super-chilled storage at -2ºC on the microbiological and organoleptic properties of cold-smoked salmon before retail display. Int J Refrig. 32 (7):1850–1857. doi:10.1016/j.ijrefrig.2009.07.001

Bhat, Z. F., Morton, J. D., Mason, S. L., & Bekhit, A. E.-D. A. (2018). Role of calpain system in meat tenderness: A review. *Food Science and Human Wellness, 7*(3), 196–204. https://doi.org/10.1016/j.fshw.2018.08.002

Bogdanović, T., Simat, V., Frka-Roić, A., & Marković, K. (2012). Development and application of Quality Index Method scheme in a shelf-life study of wild and fish farm affected bogue (*Boops boops* L.). *Journal of Food Science, 77*(2), S51–S58. https://doi.org/10.1111/j.1750-3841.2011.02550.x

Bonilla, A. C., Sveinsdóttir, K., & Martinsdóttir, E. (2007). Development of Quality Index Method (QIM) scheme for fresh cod (*Gadus morhua*) fillets and application in shelf-life study. *Food Control, 18*(4), 352–358. https://doi.org/10.1016/j.foodcont.2005.10.019

Botta, J. R. (1995). *Evaluation of seafood freshness quality*. VCH Publishers.

Chang, K. L. B., Chang, J., Shiau, C. Y., & Pan, B. S. (1998). Biochemical, microbiological, and sensory changes of sea bass (*Lateolabrax japonicus*) under partial freezing and refrigerated storage. *Journal of Agricultural and Food Chemistry, 46*(2), 682–686. https://doi.org/10.1021/jf970622c

Chen, X., Weber, I., & Harrison, R. W. (2008). Hydration water and bulk water in proteins have distinct properties in radial distributions calculated from 105 atomic resolution crystal structures. *The Journal of Physical Chemistry B, 112*(38), 12073–12080. https://doi.org/10.1021/jp803760u

Chéret, R., Delbarre-Ladrat, C., de Lamballerie-Anton, M., & Verrez-Bagnis, V. (2007). Calpain and cathepsin activities in post mortem fish and meat muscles. *Food Chemistry, 101*(4), 1474–1479. https://doi.org/10.1016/j.foodchem.2006.03.031

Church, N. (1998). MAP fish and crustaceans: Sensory enhancement. *Food Science and Technology Today, 12*(2), 73–82

Dalgaard, P., Gram, L. & Huss, H. (1993). Spoilage and shelf life of cod fillets packed in vacuum or modified atmospheres. Int. J. Food Microbiol 19: 283- 294.

Defoe, J. (2015). Development and application of quality index method scheme for fresh farmed Atlantic salmon (*Salmo salar*) fillets and whole fish. United Nations University Fisheries-Training-Programme,-Iceland-[final-project]. http://www.unuftp.is/static/fellows/document/dafoe14prf.pdf

Devahastin, S., (2010). Effect of high-pressure food processing on physicochemical changes of foods: A review. Physicochemical Aspects of Food Engineering and Processing, pp. 123–194, 2010.

Dowlati, M., Mohtasebi, S., Omid, M., Razavi, S., Jamzad, M. & de la Guardia, M., (2013). Freshness assessment of gilthead sea bream (*Sparus aurata*) by machine vision based on gill and eye color changes. J. Food Eng., 119: 277–287.

Doyle, J.P. (1989). Seafood Shelf Life as a Function of Temperature. Sea gram, Alaska sea grant marine advisory programme. No:30. 1989. 2nd printing 1995.

Duun, A. S., (2008). Super-chilling of muscle food: Storage stability and quality aspects of salmon (*Salmo salar*), cod (*Gadus morhua*) and pork," 2008. F.M. Eds. Critical Reviews in Food Science and Nutrition pp. 83-90. Florida: CRC Press.

Duun, A. S. and T. Rustad, (2008). Quality of super chilled vacuum packed Atlantic salmon (*Salmo salar*) fillets stored at- 1.4 and- 3.6ºC. Food Chemistry, vol. 106, no. 1, pp. 122–131, 2008.

Duun, A. S., and T. Rustad, (2007). Quality changes during super-chilled storage of cod (*Gadus morhua*) fillets. Food Chem. 105:1067–1107. doi:10.1016/j.foodchem.2007.05.020.

Edirisinghe, U.A.S.K., Wickramasinghe I. and Wansapala, M.A.J. (2019). Factors Affecting to Freshness Quality of Selected Tuna Fish Varieties along the Supply Chain. Challenges, Trends and Opportunities of Technical and Vocational Education and Training (TVET). International Research Symposium – University of Vocational Technology, 146-153.

Einarsson, H. 1988. Deep chilling (super-chilling, partial freezing) – a literature survey. SIKs Service series (30) Goteborg, Sweden: SIK – The Swedish Food Institute, Chalmers University of Technology.

Elisabeth, Huff-Lonergan and S. M. Lonergan, (2005). Mechanisms of water-holding capacity of meat: The role of postmortem biochemical and structural changes. Meat science, vol. 71, no. 1, pp. 194–204, 2005.

Erikson, U., E. Misimi, and L. Gallart-Jornet, (2011). Super-chilling of rested Atlantic salmon: Different chilling strategies and effects on fish and fillet quality. Food Chemistry, vol. 127, no. 4, pp. 1427–1437, 2011.

European Commission. (2005). *Commission Regulation (EC) No 2074/2005 of 5 December 2005 laying down implementing measures for certain products under Regulation (EC) No 853/2004 of the European Parliament and of the Council and for the organization of official controls under Regulations (EC) No 854/2004 and (EC) No 882/2004, derogating from Regulation (EC) No 852/2004 and amending Regulations (EC) No 853/2004 and (EC) No 854/2004*. *Official Journal of the European Union, L 338*, 27–59. https://eur-lex.europa.eu/legal-content/EN/TXT/PDF/?uri=CELEX:32005R2074

FAO (2009). Food and Agriculture Organization of the United Nations. *Salmo salar*. In *Cultured aquatic fish species fact sheets*. M. Jones (Author), V. Crespi & M. New (Eds.). https://www.fao.org/fishery/docs/CDrom/aquaculture/I1129m/file/en/en_atlanticsalmon.htm

FAO (2022). The State of World Fisheries and Aquaculture 2022. Towards Blue Transformation. Rome, FAO. https://doi.org/10.4060/cc0461en

Farber, J. M. (1991). Microbiological aspects of modified-atmosphere packaging technology: A review. *Journal of Food Protection, 54*(1), 58–70.

Feldhusen, F. (2000). The role of seafood in bacterial foodborne diseases. *Microbes and Infection, 2*(13), 1651–1660.

Froese, R., & Pauly, D. (Eds.). (2024). *FishBase: Salmo salar (Atlantic salmon)*. FishBase. https://www.fishbase.se/summary/Salmo-salar

Gallart-Jornet, L., Rustad, T., Barat, J. M., Fito, P., & Escriche, I. (2007). Effect of super-chilled storage on the freshness and salting behavior of Atlantic salmon (*Salmo salar*) fillets. *Food Chemistry, 103*(4), 1268–1281. https://doi.org/10.1016/j.foodchem.2006.10.040

Gazette, 1980. Parliament of the Democratic Socialist Republic of Sri Lanka. (2020). *Food (Fish and Fish Products) Regulations, 2020, made under the Food Act, No. 26 of 1980*. Government Gazette of the Democratic Socialist Republic of Sri Lanka.

Ge, G., Han, Y., Zheng, J., Zhao, M., & Sun, W. (2020). Physicochemical characteristics and gel-forming properties of myofibrillar protein in an oxidative system affected by partial substitution of NaCl with KCl, $MgCl_2$, or $CaCl_2$. *Food Chemistry, 309*, 125614. https://doi.org/10.1016/j.foodchem.2019.125614

Ghaly, A. E., Ramakrishnan, V. V., Brooks, M. S., Budge, S. M., & Dave, D. (2013). Fish processing wastes as a potential source of proteins, amino acids and oils: A critical review. *Journal of Microbial & Biochemical Technology, 5*(4), 107–129.

Glover, K. A., Otterå, H., Olsen, R. E., Slinde, E., Taranger, G. L., & Skaala, Ø. (2009). A comparison of farmed, wild and hybrid Atlantic salmon (*Salmo salar* L.) reared under farming conditions. *Aquaculture, 286*(3–4), 203–210. https://doi.org/10.1016/j.aquaculture.2008.09.023

Gökoğlu, N., & Yerlikaya, P. (2015). *Seafood chilling, refrigeration and freezing: Science and technology*. Wiley-Blackwell.+

Gram, L., & Huss, H. H. (1996). Microbiological spoilage of fish and fish products. *International Journal of Food Microbiology, 33*(1), 121–137. https://doi.org/10.1016/0168-1605(96)01134-8

Gunasekara, S. S., Jayasinghe, P., Jens, K., Totland, A., & Kithsiri, H. (2019). *Evaluation of historical changes in coastal fisheries of Sri Lanka using length-based indicators*. https://doi.org/10.13140/RG.2.2.22547.09764

Halvorsen, B. L., & Blomhoff, R. (2011). Determination of lipid oxidation products in vegetable oils and marine omega-3 supplements. *Food & Nutrition Research, 55*(1), Article 5792. https://doi.org/10.3402/fnr.v55i0.5792

Hamilton, M. C., Hites, R. A., Schwager, S. J., Foran, J. A., Knuth, B. A., & Carpenter, D. O. (2005). Lipid composition and contaminants in farmed and wild salmon. *Environmental Science & Technology, 39*(22), 8622–8629. https://doi.org/10.1021/es050898r

Hong, H., Regenstein, J. M., & Luo, Y. (2017). The importance of ATP-related compounds for the freshness and flavor of post-mortem fish and shellfish muscle: A review. *Critical Reviews in Food Science and Nutrition, 57*(9), 1787–1798. https://doi.org/10.1080/10408398.2014.1001489

Howgate, P. (1982). Quality assessment and quality control. In A. Aitken, I. M. Mackie, J. H. Merritt, & M. L. Windsor (Eds.), *Fish handling and processing* (pp. 177–186). HMSO.

Huss, H. H. (1988). *El pescado fresco: Su calidad y cambios de calidad* (FAO Documento Técnico de Pesca No. 348). FAO. https://www.fao.org/4/v7180s/v7180s00.htm

Huss, H. H. (1995). *Quality and quality changes in fresh fish* (FAO Fisheries Technical Paper No. 348). Food and Agriculture Organization of the United Nations. https://www.fao.org/3/v7180e/v7180e00.htm

Krakstad, J.-O., Jayasinghe, P., Totland, A., De Silva, P., Stiansen, H. E., Currie, D., Gunasekara, S., Lokubandara, U., Haputhantri, S., Rajapaksha, I., Wickramasinghe, U., Wijeratne, A., Nallawatta, S., Hapugaswatta, A., Weerasekara, R., & Kaluthotage, P. (2018). *Survey of regional resources and ecosystem off Bay of Bengal, Survey No. 2018408, Sri Lanka: Cruise report Dr. Fridtjof Nansen*. NORAD–FAO Programme (GCP/GLO/690/NOR), Institute of Marine Research (IMR), and National Aquatic Resources Research and Development Agency (NARA).

Jianadasa, B. K. K. K., Ginigaddara, P. H., & Ariyawansa, S. (2014). A comparative quality assessment of five types of selected fishes collected from retail market in Sri Lanka. *American Journal of Food Science and Technology, 2*(1), 21–27. https://doi.org/10.12691/ajfst-2-1-4

Kaale, L. D., Eikevik, T. M., Bardal, T., & Kjørsvik, E. (2013). A study of the ice crystals in vacuum-packed salmon fillets (*Salmo salar*) during super-chilling process and following storage. *Journal of Food Engineering, 115*(1), 20–25. https://doi.org/10.1016/j.jfoodeng.2012.09.006

Kaale, L. D., Eikevik, T. M., Rustad, T., & Kolsaker, K. (2011). Super-chilling of food: A review. *Journal of Food Engineering, 107*(2), 141–146. https://doi.org/10.1016/j.jfoodeng.2011.06.004

Kaale, L. D., Eikevik, T. M., Rustad, T., & Nordtvedt, T. S. (2014). Changes in water holding capacity and drip loss of Atlantic salmon (*Salmo salar*) muscle during super-chilled storage. *LWT – Food Science and Technology, 55*(2), 528–535. https://doi.org/10.1016/j.lwt.2013.10.021

Kaale, L. D., & Eikevik, T. M. (2016). Changes of proteins during super-chilled storage of Atlantic salmon muscle (*Salmo salar*). *Journal of Food Science and Technology, 53*(1), 441–450.

Kiessling, A., Espe, M., Ruohonen, K., & Mørkøre, T. (2004). Texture, gaping and colour of fresh and frozen Atlantic salmon flesh as affected by pre-slaughter iso-eugenol or CO_2 anesthesia. *Aquaculture, 236*(1–4), 645–657.

Lauzon, H. L., Margeirsson, B., Sveinsdóttir, K., Guðjónsdóttir, M., Karlsdóttir, M. G., & Martinsdóttir, E. (2010). *Overview on fish quality research: Impact of fish handling, processing, storage and logistics on fish quality deterioration* (Matís Report No. 39-10). Matís ohf. ISSN 1670-7192.

Lauzon, H., Sveinsdóttir, K., & Karlsdóttir, M. (2011). *Effect of temperature control on the efficiency of modified atmosphere packaging of cod loins in bulk* (Matis Report No. 21-11). Matis.

Lorenzo, J. M., Munekata, P. E. S., Domínguez, R., Pateiro, M., Saraiva, J. A., & Franco, D. (2018). Main groups of microorganisms of relevance for food safety and stability: General aspects and overall description. In *Innovative technologies for food preservation* (pp. 53–107). Elsevier.

Lushchak, V., (2007). Free radical oxidation of proteins and its relationship with functional state of organisms. Biochemistry (Moscow), vol. 72, no. 8, pp. 809–827, 2007.

Magnussen, O. M., Haugland, A., Hemmingsen, A. K. T., Johansen, S., & Nordtvedt, T. S. (2008). Advances in super-chilling of food—Process characteristics and product quality. *Trends in Food Science & Technology, 19*(8), 418–424. https://doi.org/10.1016/j.tifs.2008.04.005

Magnússon, H., Sveinsdóttir, K., Þorvaldsson, L., Guðjónsdóttir, M., Lauzon, H. L., Reynisson, E., Runarsson, A. R., Magnússon, S. H., Viðarsson, J. R., Arason, S., & Martinsdóttir, E. (2010). *The effect of different cooling techniques on the quality changes and shelf life of whole cod (Gadus morhua)* (Report No. 28-10). Matís ohf. ISSN 1670-7192.

Mallikage, M. (2001). *The effect of different cooling system on quality of pelagic species* [Final project]. United Nations University Fisheries Training Programme. https://www.grocentre.is/static/gro/publication/171/document/marcusprf.pdf

Marella, C., & Muthukumarappan, K. (2013). Food freezing technology. In *Handbook of Farm, Dairy and Food Machinery Engineering* (pp. 355–378). Elsevier.

Martinaud, A., Mercier, Y., Marinova, P., Tassy, C., Gatellier, P., & Renerre, M. (1997). Comparison of oxidative processes on myofibrillar proteins from beef during maturation and by different model oxidation systems. *Journal of Agricultural and Food Chemistry, 45*(7), 2481–2487.

Martinsdóttir, E. (2002). In H. Bremner (Ed.), *Safety and quality issues in fish processing* (pp. 360–378). Woodhead Publishing.

Martinsdóttir, E. (2002b). Quality management of stored fish. In H. A. Bremner (Ed.), *Safety and quality issues in fish processing* (pp. 360–378). Woodhead Publishing.

Martinsdóttir, E., Schelvis, R., Hyldig, G., & Sveinsdóttir, K. (2009). Sensory evaluation of seafood: Methods. In *Fishery products: Quality, safety and authenticity* (pp. 430–435). John Wiley & Sons.

Martinsdóttir, E., Sveinsdóttir, K., Luten, J., Schelvis-Smit, R., & Hyldig, G. (2001). *Reference manual for the fish sector: Sensory evaluation of fish freshness*. QIM Eurofish.

Massaquoi, H. (2011). *Quality changes of fish from catch to processing and during storage with focus on cooling practices and practical application in Sierra Leone* (Final project). United Nations University Fisheries Training Programme.

Matthäus, B., & Rubner, M. (2010). Oxidation of edible oils. In *Oxidation in foods and beverages and antioxidant applications: Volume 2: Management in different industry sectors* (pp. 183–238).

Milijašević, M., Babić Milijašević, J., Đinović-Stojanović, J., Vesković Moračanin, S., & Lilić, S. (2017). Changes of pH and peroxide value in carp (*Cyprinus carpio*) cuts packaged in modified atmosphere. *IOP Conference Series: Earth and Environmental Science, 85*, Article 012041. https://doi.org/10.1088/1755-1315/85/1/012041

Ministry of Fisheries and Aquatic Resources. (2021). *Fisheries statistics 2021: Annual report* (pp. 1–68). Ministry of Fisheries and Aquatic Resources. Sri Lanka.

Mørkøre, T., Vallet, J.-L., Cardinal, M., Gomez-Guillen, M., Montero, P., Torrissen, O., Nortvedt, R., Sigurgisladottir, S., & Thomassen, M. (2001). Fat content and fillet shape of Atlantic salmon: Relevance for processing yield and quality of raw and smoked products. *Journal of Food Science, 66*(9), 1348–1354. https://doi.org/10.1111/j.1365-2621.2001.tb15203.x

National Research Council. (1985). *An evaluation of the role of microbiological criteria for foods and food ingredients*. National Academies Press. https://doi.org/10.17226/372

Nielsen, D., & Hyldig, G. (2004). Influence of handling procedures and biological factors on the QIM evaluation of whole herring (*Clupea harengus* L.). *Food Research International, 37*, 975–983. https://doi.org/10.1016/j.foodres.2004.06.006

Ochiai, Y., & Ozawa, H. (2020). Biochemical and physicochemical characteristics of the major muscle proteins from fish and shellfish. *Fisheries Science, 86*(5), 729–740. https://doi.org/10.1007/s12562-020-01449-7

Oehlenschläger, J. (1997). Evaluation of fish freshness. In *Methods to determine the freshness of fish in research and industry: Proceedings of the Final Meeting of the Concerted Action "Evaluation of Fish Freshness" (AIR3CT94 2283)* (pp. 92–99). International Institute of Refrigeration.

Ólafsdóttir, G., Martinsdóttir, E., Oehlenschläger, J., Dalgaard, P., Jensen, B., Undeland, I., Mackie, I. M., Henehan, G., Nielsen, J., & Nilsen, H. (1997). Methods to evaluate fish freshness in research and industry. *Trends in Food Science & Technology, 8*(8), 258–265. https://doi.org/10.1016/S0924-2244(97)01049-2

Osibona, A. O., & Ezekiel, M. O. (2014). Chemical, sensory and microbiological changes of spotted grunter (*Pomadasys commersonnii*) under ice storage. *African Journal of Food, Agriculture, Nutrition and Development, 14*(6), 2141–2160.

Perera, W. P. D. S., Tharanga, K. A. D., Rajapakse, R. P. N. P., Himali, S. M. C., Jayasinghe, G. D. T. M., & Jianadasa, B. K. K. K. (2020). Chemical, microbiological and sensory evaluation of spotted sardinella (*Amblygaster sirm*) stored in ice. *Ceylon Journal of Science, 49*(2), 217–221. https://doi.org/10.4038/cjs.v49i2.7744

Provincial, L., Gil, M., Guillén, E., Alonso, V., Roncalés, P., & Beltrán, J. A. (2010). Effect of modified atmosphere packaging using different CO_2 and N_2 combinations on physical, chemical, microbiological and sensory changes of fresh sea bass (*Dicentrarchus labrax*) fillets. *International Journal of Food Science & Technology, 45*(9), 1828–1836.

Samaranayake, R. A. D. B. (2003). Review of national fisheries situation in Sri Lanka. In G. Silvestre, L. Garces, I. Stobutzki, M. Ahmed, R. A. Valmonte-Santos, C. Luna, L. Lachica-Aliño, P. Munro, V. Christensen, & D. Pauly (Eds.), *Assessment, management and future directions of coastal fisheries in Asian countries* (pp. 987–1012). WorldFish Center. (WorldFish Center Conference Proceedings No. 67)

Sant'Ana, L. S., Sílvia, S., & Paulo, V. P. (2011). Development of a quality index method (QIM) sensory scheme and study of shelf-life of ice-stored blackspot seabream (*Pagellus bogaraveo*). *LWT – Food Science and Technology, 44*, 2253–2259.

Seabra, L. M. J., & Pedrosa, L. F. C. (2010). Astaxanthin: Structural and functional aspects. *Revista de Nutrição, 23*(6), 1041–1050.

Shewan, J. (1962). In J. Hawthorn & J. M. Leitch (Eds.), *Recent advances in food science* (Vol. 1, p. 167).

Shewan, J. M. (1977). The bacteriology of fresh and spoiling fish and the biochemical changes induced by bacterial action. In *Proceedings of the Conference on Handling, Processing and Marketing of Tropical Fish* (p. 51). Tropical Products Institute.

Shewan, J. M., Macintosh, R. G., Tucker, C. G., & Ehrenberg, A. S. C. (1953). The development of a numerical scoring system for the sensory assessment of the spoilage of wet white fish stored in ice. *Journal of the Science of Food and Agriculture, 4*(6), 283–298.

Sivertsvik, M., Jeksrud, W. K., & Rosnes, J. T. (2002). A review of modified atmosphere packaging of fish and fishery products: Significance of microbial growth, activities and safety. *International Journal of Food Science & Technology, 37*(2), 107–127.

State Ministry of Ornamental Fish, Inland Fish & Prawn Farming, Fishery Harbour Development, Multiday Fishing Activities and Fish Exports. (2021). *Annual performance report 2021*. State Ministry of Ornamental Fish, Inland Fish & Prawn Farming, Fishery Harbour Development, Multiday Fishing Activities and Fish Exports.

Stone, H., & Sidel, J. L. (2004). Descriptive analysis. In H. Stone & J. L. Sidel (Eds.), *Sensory evaluation practices* (3rd ed., pp. 201–244). Elsevier Academic Press.

Sveinsdóttir, K., Martinsdóttir, E., Hyldig, G., Jørgensen, B., & Kristbergsson, K. (2002). Application of quality index method (QIM) scheme in shelf-life study of farmed Atlantic salmon (*Salmo salar*). *Journal of Food Science, 67*(4), 1570–1579. https://doi.org/10.1111/j.1365-2621.2002.tb10324.x

Tamarit Pino, Y. (2018). *Development of quality index method (QIM) scheme for pollock (Pollachius virens) fillets and application in a shelf-life study* [Final project]. United Nations University Fisheries Training Programme. http://www.unuftp.is/static/fellows/document/yanara15prf.pdf

Thordarson, G., Karlsdottir, M., Pedersen, R., Johannsson, M., and Hognason, A., (2015). Sub-chilling of salmon. Report summary. Matís ohf / Matis - Food Research, Innovation & Safety. ISSN 1670-7192. https://www.matis.is/media/matis/utgafa/11-15-Sub-chilling.pdf

Turan, H., Kaya, Y., & Erkoyuncu, İ. (2003). Effects of glazing, packaging and phosphate treatments on drip loss in rainbow trout (*Oncorhynchus mykiss* W., 1792) during frozen storage. *Turkish Journal of Fisheries and Aquatic Sciences, 3*(2), 105–109.

Velasco, J., Dobarganes, C., & Marquez-Ruiz, G. (2010). Oxidative rancidity in foods and food quality. In Ø. M. Andersen & K. R. Markham (Eds.), *Chemical deterioration and physical instability of food and beverages* (pp. 3–32). Elsevier.

Wang, T., Sveinsdottir, K., Magnusson, H., & Martinsdottir, E. (2008). Combined application of modified atmosphere packaging (MAP) and superchilling storage to extend the shelf life of fresh cod (*Gadus morhua*) loins. *Journal of Food Science, 73*(1), 11–19. https://doi.org/10.1111/j.1750-3841.2007.00590.x

World Bank. (2021). *Priorities for sustainably managing Sri Lanka's marine fisheries, coastal aquaculture, and the ecosystems that support them*. World Bank. https://openknowledge.worldbank.org/handle/10986/36503

Wu, C.-H., Yuan, C.-H., Ye, X.-Q., Hu, Y.-Q., Chen, S.-G., & Liu, D.-H. (2014). A critical review on super-chilling preservation technology in aquatic product. *Journal of Integrative Agriculture, 13*(12), 2788–2806. https://doi.org/10.1016/S2095-3119(14)60901-9

Zhou, G. H., Xu, X. L., & Liu, Y. (2010). Preservation technologies for fresh meat—A review. *Meat Science, 86*(1), 119–128. https://doi.org/10.1016/j.meatsci.2010.04.033

APPENDIX

QIM scheme for cultured Atlantic salmon (Salmo salar) whole fish (Martinsdóttir et al., 2001).

Quality Parameter		**Description**	**Score**
Skin	Colour/ Appearance	Pearl-shiny all over skin	0
		The skin is less pearl shiny	1
		The fish is yellowish, mainly near the abdomen	2
	Mucus	Clear, not clotted	0
		Milky, clotted	1
		Yellow and clotted	2
	Odour	Fresh sea weedy, neutral	0
		Cucumber, metal, hay	1
		Sour, dish cloth	2
		Rotten	3
	Texture	In rigor	0
		Finger mark disappears rapidly	1
		Finger mark leaves over 3 seconds	2
Eyes	Pupils	Clear and black, metal shiny	0
		Dark grey	1
		Matt, grey	2
	Form	Convex	0
		Flat	1
		Sunken	2
Gills	Colour	Red / dark brown	0
		Pale red, pink/light brown	1
		Grey brown, brown, grey, green	2
	Mucus	Transparent	0
		Milky, clotted	1
		Brown, clotted	2
	Odour	Fresh, seaweed	0
		Metal, cucumber	1
		Sour, moldy	2
		Rotten	3
Abdomen	Blood in abdomen	Blood red/ not present	0
		Blood more brown, yellowish	1
	Odour	Neutral	0
		Cucumber, melon	1
		Sour, fermenting	2
		Rotten / rotten cabbage	3
Total QIM score			**0-24**