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ADJUSTMENT OF FRESHNESS GRADING SCHEME TO COMMERCIALLY IMPORTANT SHRIMP SPECIES IN NIGERIA

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ABSTRACT

This study aimed to improve the quality and freshness grading of commercially important shrimp species in Nigeria. This was done by developing preliminary Quality Index Method (QIM) schemes for thawed brown (*Parapenaeopsis atlantica*), white (*Peneaus notialis*), and tiger (*Penaeus monodon*) shrimp species. To facilitate the implementation of quality and freshness grading practices, a shelf-life study on thawed fjord shrimp was conducted, and protocols for training assessors within the quality assurance unit of the Federal Department of Fisheries and Aquaculture were established. Thawed brown, white, and tiger shrimp species were stored on ice and photographed on storage days 0, 2, 4, 7, and 9. Changes in specific visual sensory attributes were also observed. The observed changes were used to draft a preliminary QIM scheme for each of the specie. A QIM scheme for thawed fjord shrimp was developed, and sensory, microbial, and chemical analyses were carried out on thawed fjord shrimp stored on ice for 0, 1,3, 5, 6, and 8 days after thawing to determine its shelf life. The Quality index (QI) increased linearly with storage time (R^2=0.9487). Microbial tests showed an increase in TVC and H_2S spoilage-producing bacteria as storage progressed, but the measurement of TVB-N did not show a linear correlation with storage in ice. Based on the results of the sensory and microbial tests, shrimp became unfit for human consumption on storage day 5. Based on the knowledge and experience gained, protocols for the implementation of quality and freshness grading practices and guidelines for training fishery assessors in Nigeria were established. Preliminary QIM schemes need to be finalized and implemented in Nigeria, as this would help strengthen the quality assurance of fish inspection services and improve shrimp quality management nationally.

Keywords: Quality Index Method, sensory evaluation, shelf-life, assessors, shrimp, Nigeria.

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1 INTRODUCTION

1.1 Background of study

Nigeria is a maritime country with a coastline of 853 km, 42,000[2] continental shelf area, and an exclusive economic zone of 200 nautical miles (FAO, 2007). Within the EEZ, Nigeria has exclusive rights to explore and exploit the fish, shrimp, and other natural resources (Ibeum, 2015). The coastline of the Niger Delta borders the Atlantic Ocean and contains abundant fishery resources. The coastal shelf contains rich organic debris input that arises from runoff due to frequent rains that characterize the delta basin (Figure 1). This supports rich shrimp resources on and off the coast of the Niger Delta (Sant'Ana, Soares, & Vaz-Pires, 2011).

Figure 1. Nigerian Coastline (Lynch, 2015)

Nigeria is a tropical country with abundant shrimp resources. Shrimp is mainly wild caught in Nigeria because shrimp farming is relatively new. Presently, shrimp fisheries in Nigeria are divided into artisanal shrimp (small-scale) and industrial shrimp (large-scale). Artisanal shrimping is carried out by local fishing communities using small boats (motorized and non-motorized), nets, and crude fishing implements, mainly for local consumption. Industrial shrimping involves the use of trawlers with freezing facilities, mainly for export (Etim, Belhabib, & Pauly, 2015).

Shrimp is consumed by most people worldwide. Shrimp is valued because it is delicious, easy to cook, and a good source of dietary protein and minerals. It is rich in selenium, vitamin B12, omega 3 polyunsaturated fatty acids (PUFA), and astaxanthin, a natural protein antioxidant (Ajifolokun, Basson, Osunsanmi, & Zharare, 2019). Nigeria has produced an average of 4,400 metric tons of shrimp from industrial shrimp over the last five years (Figure 2) (FDFA, 2020). Shrimp exports add approximately USD 40 million to Nigeria Gross Domestic Product annually. It is the most valuable export fishery for Nigeria, ranking number 6 among the most exported agricultural products in 2020 (National Bureau of Statistics, 2020). The major markets are France, Spain, Italy, the Netherlands, Belgium, Taiwan, and the USA.

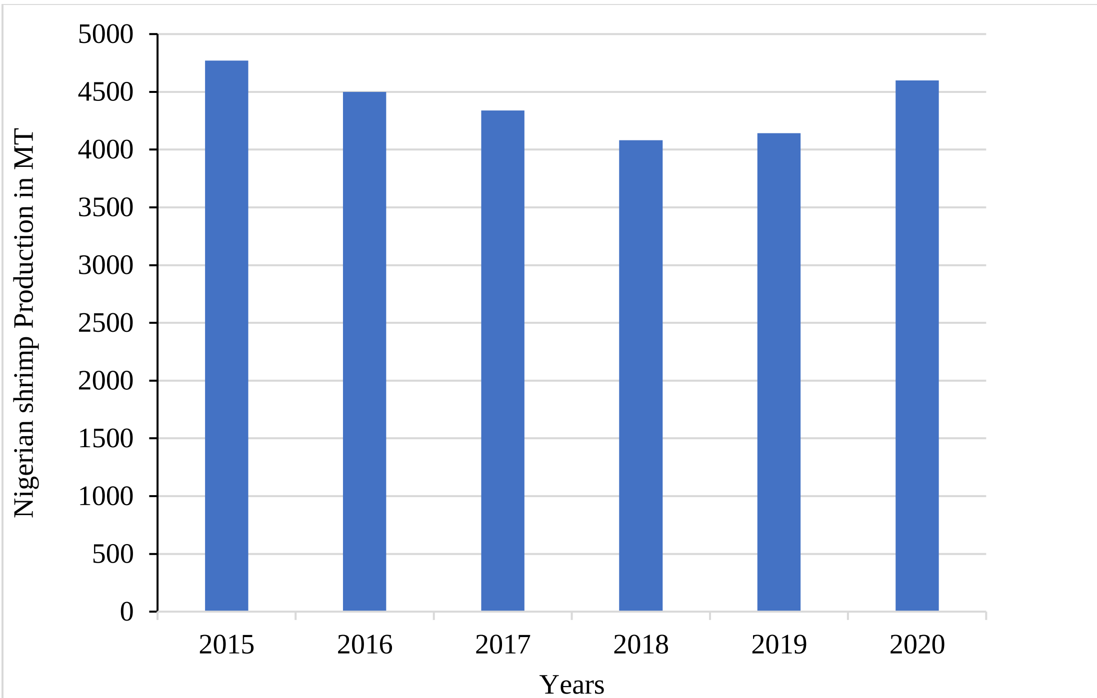


Figure 2. Nigeria industrial shrimp production 2015-2020 (FDFA, 2020)

The most commercially sought-after shrimp species in Nigeria waters are the white shrimp (*Penaeus notialis*), tiger shrimp (*Penaeus monodon)*, flower tiger shrimp (*Melicertus kerathurus*), and brown shrimp (*Parapenaeopsis atlantica*). At the shrimping ground, the catch is sorted into species, graded into sizes, and washed with clean seawater to remove dirt and debris. The shrimp are treated with sodium metabisulfite to prevent melanosis. The treated shrimp are arranged on a plate freezer tray for freezing at -30^{o}C to -40^{o}C to attain a core temperature of -20^{o}C. The frozen shrimp are packed in 2 kg rectangular packets and kept in a fish-hold at -20^{o}C to maintain the temperature for up to one to two months until the products are landed. At the landing site, frozen shrimp are offloaded into cold rooms at-20 °C, where they are stored until export. Frozen shrimp are transported in freezer containers at -20 $^{°C}$ to the seaport from where it is exported (USAID, 2002).

If shrimp is inadequately packed and stored in inadequate storage conditions, such as too high or a fluctuating temperature, it is disposed to rapid deterioration due to its high moisture and protein content. They deteriorate rapidly after capture, unless subjected to cold storage and processing, to extend their shelf life. Spoilage of seafood occurs when specific bacteria produce H_2S and certain volatile amines, such as trimethylamine (TMA), ammonia (NH3), and dimethylamine (DMA), which result in unpleasant sensory characteristics (Chan, et al., 2006).

The freshness of shrimp determines its value. Therefore, it is essential to assess freshness regularly (Sant'Ana, Soares , & Vaz-Pires, 2011). Appearance, odour, taste, and texture are sensory quality parameters that are not complex for consumers to perceive or recognize, and they tend to reject shrimp that do not have a characteristic colour and aroma. Generally, when fresh shrimp is perceived to have a fishy, rotten, or bad smell, it is rejected (Luzuriaga & Murat, 1999). Consumers and authorities understand food safety risks and call for safe handling of seafood (Ko, 2010). According to the European Union Food Safety Regulations, it is necessary to adopt measures aimed at guaranteeing that unsafe food is not placed on the market to protect human health (EC NO 178, 2002).

Sensory evaluation is the most common method used in fish inspection services to assess the freshness of seafood and quality degradation (Kim et al., 2020). Sensory analysis is critical because it plays a vital role for quality assurance. Sensory evaluation is one of the quality control measures that seafood companies use to guarantee that the products meet the expectations of buyers and regulatory agencies. Seafood buyers use sensory evaluation to ensure that the products meet their expectations, and government agencies use sensory evaluation along with microbiological and chemical analyses to ensure that seafood products meet the required standards. In Nigeria, the Quality Assurance Division of the Federal Department of Fisheries is the competent authority responsible for assessing shrimp quality throughout the value chain.

The Quality Index Method (QIM) (Bremner, 1985) is a structured scaling method used to evaluate the sensory attributes of fish and shellfish. It is a fast and inexpensive method to assess seafood freshness without the need for equipment. Only trained assessors are needed, and the process does not destroy the wholesomeness of the overall sample (Rehbein & Oehlenschlager, 2009; Daiella, Eliane, & Monica, 2013). Panellists are trained to increase their sensory sharpness, familiarize themselves with the basic knowledge of procedures used in sensory evaluation, and develop their ability to identify and describe sensory stimuli related to seafood products. The QIM scheme for freshness grading of fjord shrimp (*Pandalus borealis*) a commercially important shrimp in Iceland (Martinsdottir, Sveinsdottir, Luten, Schelvis, & Hyldig, 2001), will be modified to suit the characteristics of commercially important shrimp in Nigeria.

1.2 Problem statement

Shrimp spoilage is of significant concern to shrimp processing industries and shrimp farmers (Sahna, Xavier, Sukham, Bhusan, & Nagalakshmi, 2018). In Nigeria, sensory knowledge for determining shrimp freshness is lacking at every step of processing on board the vessel, at landing sites, markets, processing facilities, and storage facilities (frozen storage). There is no grading scheme to guide inspectors throughout the value chain. Within the quality assurance inspection service, there is no established protocol for sensory assessment or the selection and training of inspectors for the sensory evaluation of shrimp freshness.

1.3 Rationale

One primary concern of stakeholders is how to evaluate the freshness of shrimp (frozen and thawed). It is important to evaluate shrimp spoilage characteristics in order to improve shrimp quality management (Dabade et al., 2015). This study will be beneficial to all shrimp stakeholders, including the Federal Department of Fisheries, Quality Assurance Inspectors, shrimping companies, processors, and marketers. It will provide a basis for the development of tools and methods to improve shrimp quality management (Hyldig & Petersen, 2005). In addition, this study will increase the confidence level of buyers/consumers, thereby opening more markets for commercial shrimp in Nigeria, which will in turn create employment, boost production, and increase Gross Domestic Product. This study is also important because sensory evaluation of seafood is required by the European Union (EU, 2004).

1.4 Goal

The goal of this project was to improve the quality and freshness grading of commercially important shrimp species in Nigeria for better quality management.

1.4.1 Objectives

To achieve this goal, the following specific objectives were undertaken:

- Training of assessors to use the current QIM scheme for fresh whole shrimp (Fjord shrimp).
- Adapting the current QIM scheme for fresh whole shrimp to the characteristics of frozen/thawed whole shrimp.
- Shelf-life study of thawed whole fjord shrimp.
- Drafting an adjusted QIM scheme for three major species (Tiger, White and Brown) based on photos from the landing site in Nigeria.
- Establish protocols for training assessors working within the fisheries quality assurance unit of the Federal Department of Fisheries and Aquaculture in Nigeria.
- Plan for finalization of the QIM scheme in Nigeria and implementation of sensory evaluation of shrimp freshness.

2 LITERATURE REVIEW

2.1 Overview of the Nigerian Shrimp Industry

Shrimping in Nigeria's mangroves started as far back as 1950 and was operated mainly by foreign fishing vessels. However, the discovery of a large stock of shrimp in Nigeria's territorial waters in 1961, which could sustain the shrimp trawling industry, led to the commercialization of shrimp. Prior to this, shrimp were classified as a bycatch of commercial fishing, whereas the major fish stock sought by foreign trawlers was finfish. This changed in 1989 as the shrimp value increased because of the devaluation of the naira. This led to increased shrimp capture by commercial trawlers, who saw it as an added opportunity to earn more foreign exchange (USAID, 2002).

Industrial shrimp fisheries dominate Nigeria's shrimp production and are conducted within territorial waters beyond 5 NM from the shore. It involves the use of bottom trawl with a cod-end stretched mesh square nets of 44 mm, as stipulated by law. Typically, the recommended trawler size for shrimp must not exceed 130 gross tonnage and 23.4 meters in length. In addition, each trawler has a Turtle Exclusion Device (TED) and By-catch Reduction Devices (BRDs) for the escape of turtles and juveniles of non-target species (FGN, 1992). Currently, Nigeria has 144 licensed shrimp trawlers.

The major shrimping grounds are off the Escravos, Forcados, Ramos, and Brass Rivers along the continental shelf (Figure 3). Shrimping is done all year round, but catch is usually high between May and October for white shrimp, and November to April for brown shrimp (Etim, Belhabib, & Pauly, 2015). The shrimp industry is based in Lagos. The Federal Department of Fisheries and Aquaculture (FDFA), under the Federal Ministry of Agriculture and Rural Development, is the competent authority that licenses shrimp vessels and assures the quality and safety of seafood sold locally and to international markets. The shrimp are sold frozen, either as PUD (peeled but undeveined shrimp tails, that is, with the gut left in), head-on, or head-less.

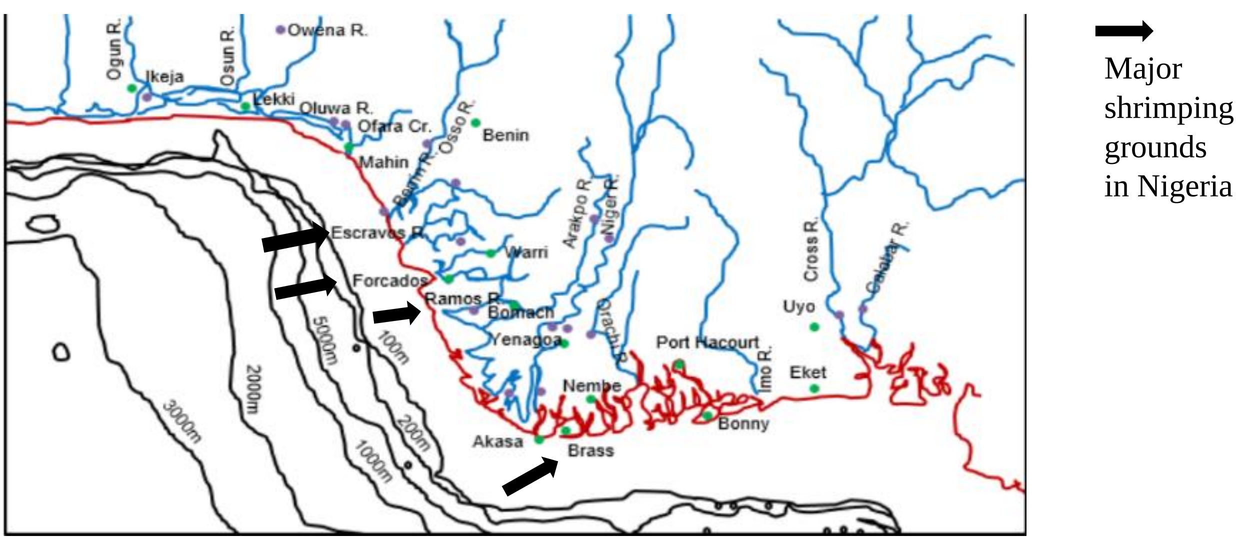


Figure 3. Map showing the major shrimping grounds in Nigeria (Francis & Oghenekevwe, 2019)

2.2 Commercially important shrimp species in Nigeria

2.2.1 Wild Tiger shrimps (Penaeus monodon)

Penaeus monodon (Figure 4) is commonly known as the 'giant tiger prawn', 'Asian tiger shrimp' and 'black tiger shrimp' (FAO, 2010). In Nigeria, it is popularly referred to as 'Wild Tiger'. Large populations of this species are found in West Africa (Fuller, et al., 2014; Knott, Fuller, Benson, & Neilson, 2019).

Penaeus monodon has adapted to many different environments (Motoh, 1985). Juveniles are generally found in sandy estuaries and mangroves, and during adulthood, they move to deeper waters (0-110 m) to live on muddy and rocky bottoms (FAO-FIRA, 2019) and are nocturnal in the wild. During the day, it burrows into the substrate, and at night it comes out to feed. It feeds on small crustaceans, algae, mollusks, detritus, and polychaete worms (Kongken, 2019). Mating occurs at night, and they produce up to 800,000 eggs (Motoh, 1985). Males are slightly smaller than females. They weigh 100-170 gm and are usually 20-25 cm long. Females are usually 25-30 cm long but can reach approximately 33 cm and weigh 200-320 g (FAO, 2010). In Nigeria, this species is caught using bottom trawl nets on a fishing vessel, and it accounts for 48% of the total catch of Nigeria's industrial shrimp fisheries between 2015 and 2020 (FDFA, 2020). It is high in carbohydrates, crude protein, fat, crude fibre, ash, and moisture. The mineral content composition for 100 g of fresh *P. monodon* is 128.8 mg magnesium, 233.7 mg phosphorous, 142.2 mg calcium and 117.3 mg sodium (Edah, Adeyemi, & Pedro, 2016).

Figure 4. External biology of Penaeus monodon (Sivakumar, Udeni , & Chitravadivelu, 2015)

2.2.2 White Shrimps (Penaeus notialis)

Penaeus notialis (figure 5) is globally known as southern pink shrimp; however, in Nigeria, it is commonly called white shrimp. It is a marine species distributed in the eastern Atlantic (West African coast from Mauritania to Angola), Western Atlantic (from Cuba to the Virgin Islands), and Atlantic coast of Middle and South America (from South Mexico to Brazil). This species is usually found at depths of 3-100 m, and can rarely be found as deep as 700 m. They live in bottom mud, sandy mud, and sandy patches among rocks in the marine environment. Juveniles are mostly found in the estuaries. *P. notialis* grows to a maximum total length of 175 mm (males) and 192 mm (females) (Holthuis, 1980), and it is an important commercial species in West Africa.

Penaeus notialis is high in carbohydrates, crude protein, fat, crude fibre, ash, and moisture. The mineral content composition for 100 g of fresh *P. notialis* is 174.8 mg magnesium, 242.3 mg phosphorous, 134.8 mg calcium and 199.2 mg sodium (Edah, Adeyemi, & Pedro, 2016). It accounted for 25% of the total catch of Nigeria's industrial shrimp fisheries between 2015 and 2020 (FDFA, 2020)

Figure 5. White/ Southern pink shrimp (Zacharie, 2011)

2.2.3 Brown shrimps (Parapenaeopsis atlantica)

Parapenaeopsis atlantica (Figure 6) is a tropical shrimp commonly known as the 'brown shrimp' in Nigeria. It accounted for 21.3% of the total catch of Nigeria's industrial capture fisheries between 2015 and 2020 (FDFA, 2020) and is distributed in the Eastern Atlantic and Western Indian Ocean along the West African coast from Senegal to Angola and around the Southern Cape to Mozambique. It is a benthic species found within a depth range of 1-60 m, usually 10-40 m. It is found in bottom mud or sandy mud marine and estuarine environments with temperatures of not less than 16 °C (Holthuis, 1980). This species grows to a maximum length of 12 cm for male and 17 cm for female (Fischer, Bianchi, & Scott, 1981). It has a maximum published weight of 23 g (Udoinyang, Iheukwumere, Amali, & Ukpatu, 2016). One hundred grams of this species contains 99 Kcal/410 KJ, protein 23%, carbohydrates <0.5% carbohydrates, and < 0.7% fat (Primstars, 2022).

Figure 6. Brown shrimps (*Parapenaeopsis atlantica*) (Primstars, 2022)

2.3 Commercially important shrimp species in Iceland

2.3.1 Deep water shrimps (Pandalus borealis)

Pandalus borealis (figure 7) is a caridean shrimp species that is found in cold parts of the northern Pacific and northern Atlantic Oceans (FAO, 2010) although the former population is now most often regarded as a separate species *Pandalus eous* (Fransen, 2019). They are commonly referred to as the 'northern shrimp' by the Food and Agriculture Organization. Other common names are deep-water prawns, cold-water prawns, nordic shrimps, and marine shrimps (FAO, 2010).

Pandalus borealis normally lives on soft muddy bottoms at depths of 20-1330 m (FAO, 2010) in temperate waters of 0-8^{0}C (Muus, Nielson, Dahlstrom, & Nystrom, 1999), although it has been recorded from 9-1450 m (Palomares & Pauly, 2019). This species grows well in saline waters (Eurofish Magazine, 2020) and is distributed in the United States, southern and eastern Greenland, Canada's eastern seaboard, Iceland Svalbard, Norway, and North Sea. This species has a lifespan of up to eight years. Males can reach a length of 12 cm, while females can reach 16.5 m long. Due to the sex of the shrimp and temperature changes in the water, the size of the species varies at different ages (Fransen, 2019). The spawning season for northern shrimp begins in late summer, usually offshore. By early fall, females begin extruding their eggs into their abdomen. They later move inshore to hatch eggs in winter.

Figure 7. Deep water shrimps (*Pandalus borealis*) (WoRMS, 2011)

2.4 Shelf life and spoilage of shrimps

Shelf life is defined as the period during which a food product is suitable for human consumption. With shrimp, it represents the time from catch until it becomes unfit and is no longer safe for human consumption (Martinsdottir, 2002). Shrimps are highly perishable after being harvested, and unlike other shellfish (lobsters and crabs), shrimp cannot be kept alive until processing because they die immediately after being caught. Bacteria from the environment as well as the mud trawled up with them are the primary cause of shrimp contamination (Adams & Moss, 2000).

Spoilage occurs because of metabolic changes in food that make it less palatable, undesirable, and unfit for human consumption. Alterations in taste, changes in smell, and appearance are usually accompanied by spoilage (Doyle, 2007). The Non-Protein Nitrogen (NPN) content of shrimp is relatively high. The NPN fraction consists of nucleotides, free amino acids, and other compounds that are suitable substrates for the growth of microbes that cause spoilage. Susceptibility of shrimp to spoilage is increased because of higher water activity and neutral pH (Sahna, Xavier, Sukham, Bhusan, & Nagalakshmi, 2018) Sensory spoilage occurs due to protein decomposition and metabolites formation from the growth of microorganisms.

The organisms responsible for shrimp spoilage differ according to species, geographic origin, processing methods, and storage conditions (Adams & Moss, 2000). Spoilage in shrimp can be classified as microbiological, chemical, or enzymatic. Spoilage reduces the quality of shrimp by the giving off of odour, deteriorating flavour, changing colour, and changing texture (Dabade, et al., 2015). Of these sensory attributes, the most offensive is spoilage odour, which is due to the combination of short-chain fatty acids, sulphides, and amines (Du, Chai, Guo, & Lu, 2015).

2.4.1 Microbial spoilage

The spoilage of seafood products is primarily caused by microorganisms (Gram and Dalgaard, 2002). Several factors, such as the shrimp species, environment, salinity and temperature of the water, and geographic location, influence the bacterial flora of the shrimp (Dabade, 2015). Bacteria hydrolyse proteins, especially structural proteins, into peptides and amino acids, which leads to changes in the physicochemical properties of the flesh, such as colour, texture, water holding capacity, and moisture (Zhuang, et al., 2019). Microbial decomposition of amino acids containing sulphur, aromatic amino acids and branched-chain amino acids produces volatile organic compounds (VOC) which produces off odour (Marilley & Cassey, 2004)

Total volatile base nitrogen (TVB-N), which includes trimethylamine (TMA), dimethylamine (DMA), and ammonia (NH3), is the most widely used parameter for measuring the microbial spoilage of seafood (Chan et al., 2006). The ability to produce hydrogen sulphide (H_2S) and reduce trimethyl oxide (TMAO) is a characteristic of spoilage bacteria. Total Viable Count (TVC) using the pour plate method with different peptone-rich substrates containing ferric citrate have been used to detect H_2S producing bacteria (Gram & Huss, 1996). Plate count techniques have revealed that, in general, tropical shrimp carry higher initial numbers of bacteria, 10^5 -10^6 CFU/g, than cold-water species, 10^2 -10^4 CFU/g (ICMSF, 2005).

Gram-positive bacteria dominate the initial bacterial flora of shrimp from shallower waters (4-15 m), whereas *Pseudomonas* species are dominant in shrimp from deeper waters (100 m). The dominant microbes in shrimp stored on ice are mainly *Pseudomonas spp.*, whereas the main bacteria in shrimp stored in ice slurry are *Shewanella putrefaciens,* which are isolated as black colonies. The nature of this initial bacteria floral influences the shelf life and sensory properties of shrimp (Chinivasagam, Bremner, Thrower, & Nottingham, 2010).

2.4.2 Autolytic spoilage

Autolysis is the result of enzymatic actions in shrimp. Enzymes degrade the chemical components of shrimp, such as lipids, proteins, and carbohydrates, thereby altering the flavour, colour, and odour of the shrimp. Autolysis is the major cause of blackspot formation on shrimp. Amino acid degradation results in the formation of melanin pigment which gives the shrimp poor appearance (Huss, 1995)

2.4.3 Chemical spoilage

Chemically, shellfish contains lipids, carbohydrates, proteins, moisture, and vitamins in proportions that produce a characteristic structure, texture, flavour, odour, and nutritional value. Chemical degradation occurs because of oxidative rancidity. An increase in temperature favours chemical degradation. Proteins are degraded into hydrogen sulphide by bacteria. Fermentative organisms degrade carbohydrates into acids, alcohols, and gases, whereas lipolytic microorganisms degrade lipids into fatty acids. All these chemical changes results in loss of odour, colour, and texture of shellfish (Huss, 1994)

2.5 Sensory Evaluation of shellfish

Sensory evaluation is an experimental approach used to stimulate, measure, assess, and interpret the sensory responses of food products. It measures consumers' feedback of food which are discerned by sight, smell, touch, and taste (Stone, Bleimbaum, & Thomas, 2020). It is a subjective evaluation that relies largely on the opinions of selected individuals. Assessors of sensory evaluation must be trained to obtain reliable results (Martinsdottir, 2010). In the fish sector and fish inspection services, one of the most important methods for assessing freshness and quality is sensory evaluation (Sant'Ana, Soares , & Vaz-Pires, 2011)

One of the main qualities of raw shrimp is its odour. Shrimp is said to be unfit for consumption or rejected if it is observed to have a fishy, spoilage, or putrid odour. The occurrence of black spots, off odour formation, and meat firmness are the main sensory and biochemical quality features because of their close correlation with decreases in freshness. Spoilage odour and meat softening due to rapid freshness drop are caused by lipid and protein degradation by microorganisms (Du, Chai, Guo, & Lu, 2015). Discoloured and occurrence of black spots is commonly caused by the activity of tyrosinase during storage (Mu, Fang, Mao, Gao, & Chen, 2012). The heads of frozen shrimp have been reported to appear darker when thawed due to the effect of deep-freezing temperatures (Lei Jin, Guofang, Peipei, Jie, & Xiaojun, 2018). Other sensory quality parameters include shell colour, adhesion of the head and body, state of internal organs, and meat colour. It is crucial to promptly evaluate freshness because it is directly related to the value of a commodity.

2.5.1 Quality Index Method (QIM)

The Quality Index Method (QIM) is a leading objective sensory tool that is currently used to assess the freshness (and thus quality) of seafood (Ainaz, 2019; Esteves & Anibal, 2021), and is based on sensory attributes that are considered important and well defined, such as odour, colour, and texture, which are specific for each shrimp species. The QIM was developed at the Tasmanian Food Research Division Unit of the Commonwealth Scientific and Industrial Research Organization (CSIRO) of Australia in the late 1970s and the early 1980s (Esteves & Anibal, 2021).

The QIM technique is a practical rating system in which the sensory parameters of shrimp are inspected, and demerit points based on a well-defined description of attributes are recorded. These characteristics are scored using a 0-3 demerit points' scale, and the sum is indicated as the quality index (QI) (Ainaz, 2019). The higher the score for each freshness attribute, the higher the spoilage of the shrimp, which indicates the specimen's "lack of freshness," whereas low scores indicate superior quality. The QIM scheme is based on the assertion that assessors can easily detect changes in a product, although they are unable to judge the degree of perfection (Hyldig, Martinsdottir, Sveinsdottir, Schelvia, & Bremner, 2010). However, the QIM scheme is usually associated with microbiological and physiochemical analysis to estimate the shelf life of the species (Lanzarin, et al., 2016)

One of the objectives of developing a QI is to establish a linear correlation between sensory quality (expressed as QI) and storage time in ice, which makes it possible to estimate shelf life (Le et al., 2017). As the temperature and storage period of seafood increase, the chemical, biochemical, and microbiological test parameters also increase (Bernadi et al., 2010; Bernadi, Marsico, & Freitas, 2013). The advantage of using QIM is that it maintains sample integrity and estimates the period during which seafood stored at chilling is safe for consumption (Sveinsdottir et al., 2002). Separate schemes exist for various shrimp species based on spoilage indicators and quality standards for each species.

Quality Index (QI) development occurs in three stages: two steps for training the assessors and one step for validation. In the first training step, the decay process and seafood attributes that changed during storage are observed and described by assessors (Ritter et al., 2016). The changes are noted in a preliminary QI protocol and each set of observations is given a score from 0-3 (Guillerm, et al., 2006). The second training step is carried out to confirm the listed observations and suggest changes to the preliminary QI protocol (Sant'Ana, Soares, & Vaz-Pires, 2011), while the third and final step is the validation of the QI protocol (Le et al., 2017). QIM scheme is available for whole, unfrozen fjord shrimp (Martinsdottir, Sveinsdottir, Luten, Schelvis, & Hyldig, 2001).

3 MATERIALS AND METHODS

3.1 Experimental Design

The experiment was carried out in two parts (Figure 8): I) Frozen/thawed whole fjord shrimp was used to train the panellists on the procedure of adapting the available QIM scheme for fjord shrimp (Martinsdottir, Sveinsdottir, Luten, Schelvis, & Hyldig, 2001) to thawed fjord shrimp in a shelf life study, and II) photos of three Nigerian shrimp species thawed and stored in ice were used to help develop preliminary QIM schemes for these species, referred to as white shrimp (*Penaeus notialis*), tiger shrimp (*Penaeus monodon*), and brown shrimp (*Parapenaeopsis atlantica).*

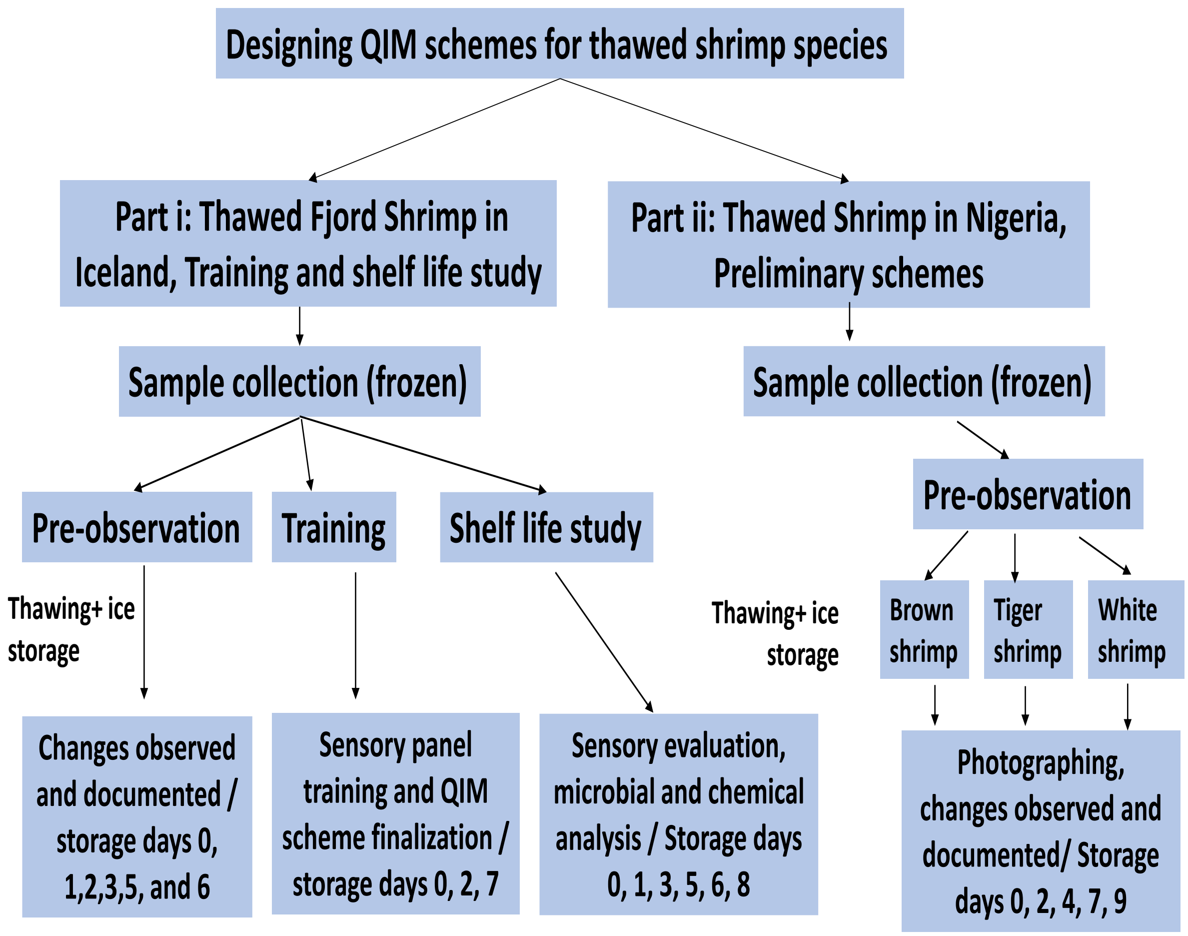


Figure 8. Experimental design of the study

3.2 Raw materials

Frozen whole fjord shrimp were purchased from Kampi in Iceland. The sample was packed in a carton box insulated with a plastic layer and transported to the Matis Laboratory. Frozen whole fjord shrimp arrived at the Matis laboratory on the 19th of December 2021. The sample was stored at -18 $^{\circ}C$. The shrimp were thawed at ambient temperature and stored at temperatures between 0-4^{o} for training purposes and shelf-life studies.

Whole *Penaeus notialis*, *Penaeus monodon* and *Parapenaeopsis atlantica* shrimp species caught with bottom trawl within Nigeria's territorial waters and frozen on-board vessel were bought at an approved landing site in Lagos, Nigeria. Photographs of frozen samples were taken from the landing site. Frozen samples were transported in an ice box to the Nigerian Fisheries Laboratory (NFL), Lagos, Nigeria. Each species was thawed separately at ambient temperature, and photographs of the thawed shrimp samples were taken. Thawed samples of each species (15 shrimp in each group) were stored separately at 0-4^{o}C in ice for 10 days and pre-observed for changes in odour. Photographs of each thawed species were taken on storage days 0, 2, 4, 7, and 9. The photographs were studied at Matis, Reykjavik, Iceland, and utilized during the adaptation of a preliminary QIM scheme for each of the three species.

3.3 Development of a Preliminary QIM scheme (for thawed fjord shrimps)

On the 20th of December 2021, 15 shrimps were removed from the frozen samples and thawed at ambient temperature in the sensory evaluation laboratory for 40 minutes (Figure 9). The thawed shrimp were arranged on a white rectangular board, placed under white, fluorescent light, and pre-observed for sensory changes using the QIM scheme for whole fjord shrimp (Martinsdottir et al., 2001). The thawed shrimp samples were observed for changes in the colour of the head, roe and hue colour, odour, homogeneity, and dryness of the body. At the end of the pre-observation period, the sample was wrapped in plastic, placed on ice, and stored at 0-2^{o}C (Figure 10). This pre-observation process was repeated on storage days 1, 2, 3, 5,6 and 7. Based on the observed changes, the QIM scheme for whole fjord shrimp (Martinsdottir et al., 2001) was slightly modified for training panellists. This pre-observation was conducted by three participants.

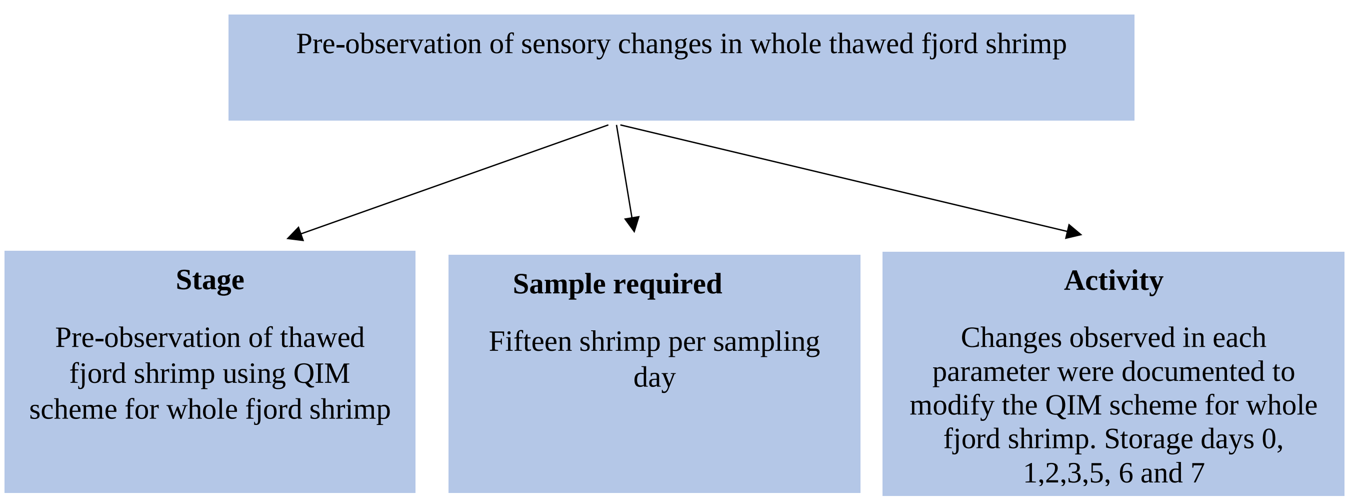


Figure 9. Experimental design for the development of a QIM scheme for thawed fjord shrimp.

Figure 10. Storage of thawed fjord shrimp on ice during pre-observation.

3.4 Training of panellists

The preliminary scheme developed for thawed fjord shrimp was used to train panellists. Six panellists were trained according to international standard guidelines (ISO 8586, 2012) at the sensory laboratory using the preliminary scheme developed during the pre-observation. Samples of thawed shrimp on storage days 0, 2, and 7 were used for this training. Each sample consisted of 15 shrimp that were placed on a sensory table under bright light. A written note was placed close to each sample to indicate the storage day for easy identification (Figure 11). The training was conducted in two sessions, providing time to modify the preliminary scheme.

During the first training session, the panel leader explained how to use the pre-liminary scheme and the meaning of each quality attribute. The panellists evaluated the samples and discussed the evaluation and description of sensory attributes among themselves and the panel leader. At the end of the first training session, the panellists provided their comments and suggested the changes they considered needed for the preliminary scheme.

Prior to the second training session, changes were made to the preliminary scheme. This modified scheme was then used by the panellists to evaluate the same samples. Some further suggestions and corrections were also made during this session. At the end of training, changes were made to the preliminary scheme. Some parameters were changed to suit the characteristics of the samples better, and a QIM scheme for thawed fjord shrimp was developed.

Figure 11. Training of panellists in sensory evaluation of thawed fjord shrimp.

3.5 Development of a preliminary QIM scheme for white, tiger and brown shrimp

During the pre-observation period in Nigeria, changes in odour were observed and documented by a research assistant. The documented odour changes and photos of the shrimp samples were sent from Nigeria to the Matis Laboratory, Iceland. The photos were carefully observed for changes in general appearance, head colour, and colour of the legs and tail that occurred in the three different species throughout the storage period (Figure 12). Based on the observed changes, the QIM scheme for whole fjord shrimp was adjusted to develop preliminary QIM schemes for white (*Penaeus notialis)*, tiger (*Penaeus monodon)*, and brown (*Parapenaeopsis atlantica)* shrimp species that can be used to train panellists in the sensory laboratory of the Federal Department of Fisheries, Lagos, Nigeria.

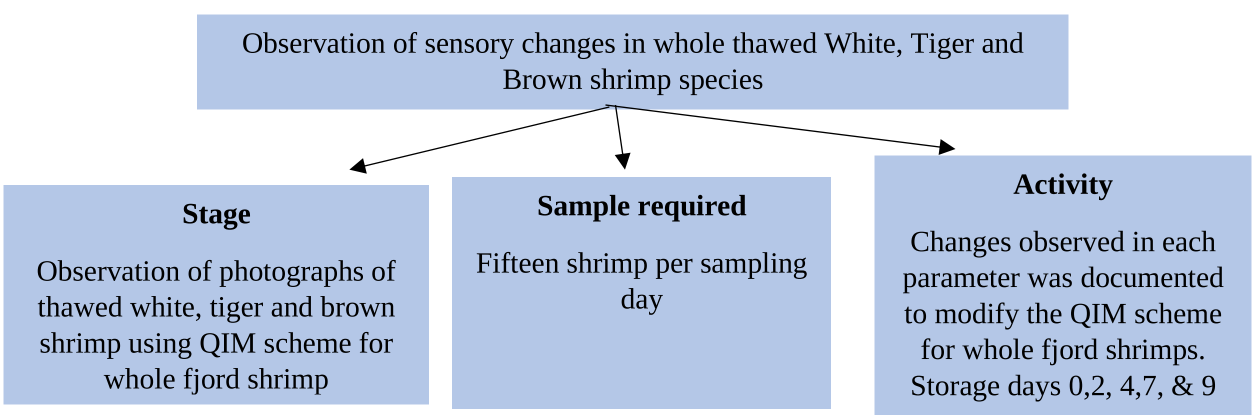


Figure 12: Experimental design for whole thawed whole White, Tiger and Brown shrimp species

3.6 Shelf-life study

Sensory evaluation and microbial and chemical analyses were performed on thawed fjord shrimp samples stored in ice on storage days 0,1,3,5,6 and 8 to determine their shelf life.

3.6.1 Sensory evaluation

Sensory evaluation was carried out on thawed fjord shrimp stored on ice using a modified QIM scheme. Six trained and three untrained panellists evaluated samples on different storage days in two sessions to determine the QI of each sample. During the first sensory session, samples on storage days 0, 3, and 6 were presented in triplicate to the panellists. During the second session, samples from storage days 1, 5, and 8 were presented in triplicate to the panellists. Each sample contained 10 shrimp. The storage days of the samples were not disclosed to the panellists, as each sample was coded with a random 3-digit number and placed on a clean table under bright light. The sensory attributes evaluated were odour, roe colour and colour of the head/abdomen/tail. Each panellist evaluated each sensory attribute described in the QIM scheme for thawed fjord shrimp and scored samples accordingly. This sensory evaluation was performed in the sensory evaluation laboratory at Matis (Figure 13).

Figure 13. Panellists during sensory evaluation of thawed fjord shrimp.

3.6.2 Microbial analysis

Ten shrimp samples at storage days 0, 1, 3, 5, 6, and 8 were taken to the microbiological laboratory at Matis before the commencement of the sensory evaluation. At the microbiological laboratory, 25 g of each sample was minced in a blender. The minced sample was placed in a stomacher bag and mixed with 180 g of Maximum Recovery Diluent (MRD; Oxoid, UK) in a stomacher for one minute. Successive 10-fold dilutions were performed, as required. The dilutions were pipetted into Petri dishes, and melted iron agar was poured into the Petri dish, mixed, and allowed to solidify.

All plates were incubated at 17 °C for 5 days. Counts of all colonies (both white and black) provided the total count and counts of black colonies as the number of H_2S-producing bacteria (NMKL184, 2006). The counts of all colonies were expressed as a logarithm of the number of colony-forming units per gram (Log Cfu/g).

3.6.3 Chemical analysis

Shrimp samples on storage days 0, 1, 3, 5, 6, and 8 were analysed for total volatile base nitrogen using the Kjeldahl distillation method (Malle & Poumeyrol, 1989). An aqueous solution of 7.5 % (100 mL) was added to 50 g of the minced sample and mixed in a blender for 60 seconds. A solution of boric acid and indicators was placed under the condenser compartment of the distillation unit, whereas the sample solution was placed under the heated compartment of the unit. Distillation was performed for 4 min, which turned the boric acid solution green. The green boric acid solution was titrated with 0.0395N aqueous H2SO4 until it turned pink and neutralization was complete. The result was calculated as mg N/100 g using the following formula:

$$\text{mgN/100g:} \quad \frac{14\text{mg} * a * b * 300}{25\text{ml}}$$

a: ml of H_2SO_4 acid
b: Normality of Sulphuric acid

3.7 Data Analysis

The data obtained from this study were analysed using Microsoft Excel statistical tool pack 2016. This statistical tool pack was used to generate linear equation graphs and regression correlations for QIM, TVBN, and TVC against the storage time. The microbial counts of the samples were converted into log values.

4 RESULTS

4.1 Development of a QIM scheme (for thawed fjord shrimp)

4.1.1 Preliminary scheme

Thawed fjord shrimp stored in ice were first pre-observed for six days to modify the existing QIM scheme for whole fjord shrimp (Martinsdottir, Sveinsdottir, Luten, Schelvis, & Hyldig, 2001). During this pre-observation period, the changes observed were similar to the QIM scheme for fresh/unfrozen whole fjord shrimp, except for the darker appearance of the heads and the appearance of a slightly brownish hue around the appendages on storage day 2 in frozen/thawed shrimp. A maximum of 11 points was recorded for the maximum quality index (Table 1).

Table 1. Preliminary Quality Index Method (QIM) scheme for thawed fjord shrimp.

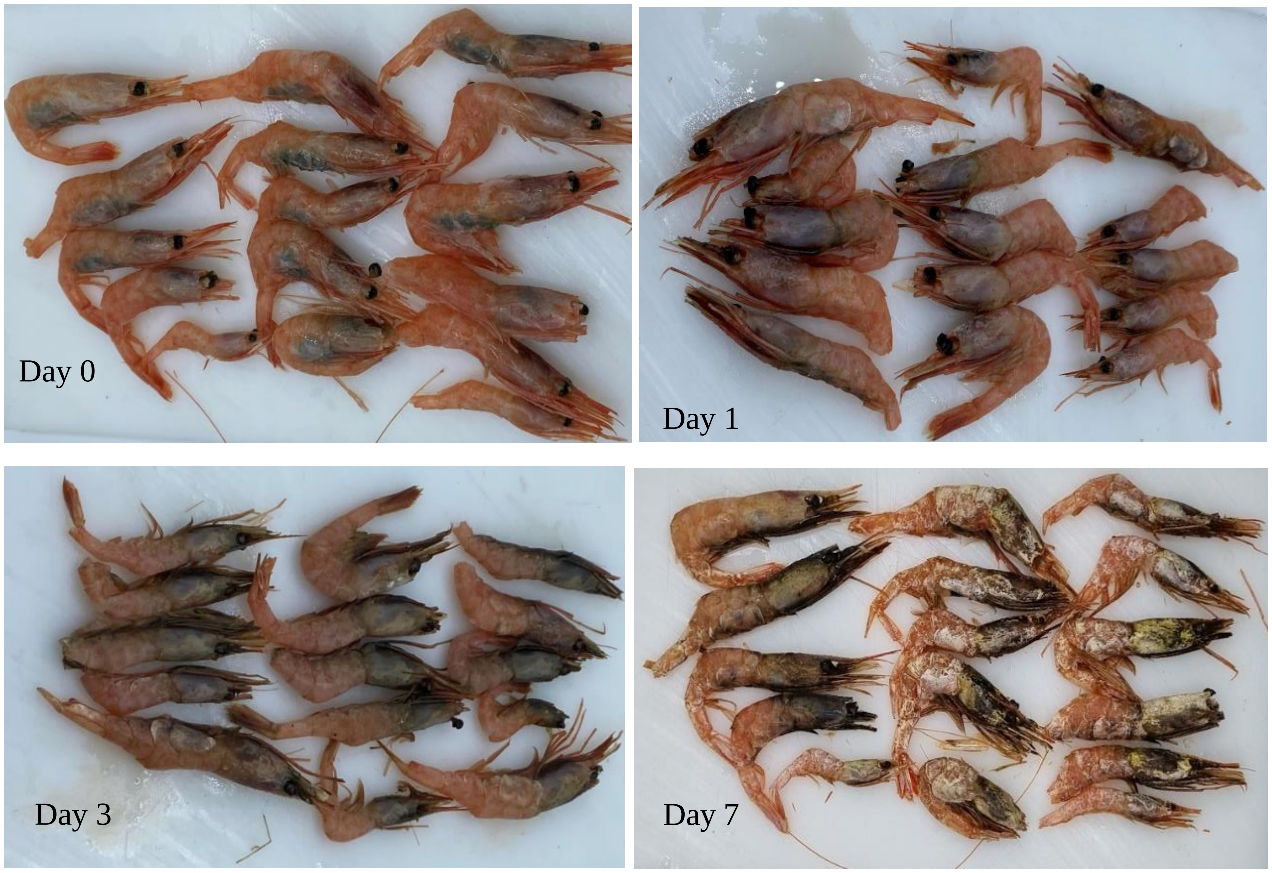
Quality parameter		**Description**	**Score**
Whole shrimp	**Dark in the head**	None or few (0-25%)	**0**
		Some (25 - 50%)	**1**
		Many (50-75%)	**2**
		All (75-100%)	**3**
	Colour	Rather pale pink, red, bright/clear colour	**0**
		Pale pink	**1**
		Brownish, Yellowish	**2**
		Yellow, green-, greyish discolouration	**3**
	Odour	Fresh, sea weedy	**0**
		Faint odour, reminds of tar	**1**
		faint ammonia odour, fishy, sour	**2**
		Obvious ammonia odour, sour, putrid	**3**
Roe	**Roe colour**	Copper green	**0**
		Discoloured, faded, yellowish	**1**
		Dark	**2**
Quality Index			**0-11**

4.1.2 Finalization of the QIM scheme

During the training sessions, the panellists observed that the heads of all the shrimp samples appeared dark with different discolorations to varying degrees, so the panellists suggested that the description for the parameter “dark in the head” be changed to better describe how dark and discoloured the head appeared. The panellists also suggested that the quality parameter “colour” be made more specific to show what part of the shrimp's body it refers to. It was observed that when the colour of the shrimp turned pale pink, it became more matt. The panellists also observed that none of the samples had sour odour and as such suggested that "sour" be removed from the odour description. The appearance of the samples changed from bright pink with greenish roe on storage day 0, to pale pink, slight brownish/yellowish discolorations (head) on storage day 2, to brown/yellowish/greenish colour in most parts of the heads and some part of the abdomen with faded roe on storage day 3, and finally to dark, yellow, green, greyish discolorations extending from the head to the abdomen and tail with dark/yellow roe on storage day 7 (Figure 14). At the end of the training sessions, necessary changes and adjustments were made to the preliminary scheme to develop a QIM scheme for thawed fjord shrimp (Table 2).

Table 2. Quality Index Method (QIM) scheme developed for thawed fjord shrimp.

Quality parameter		**Description**	**Score**
Whole shrimp	**Dark in the head**	None or few, small part of the head, slightly brownish	**0**
		Majority of the shrimp has brownish or slightly yellowish discolouration in all or most of the head	**1**
		Most or all the shrimp have brown, yellowish, greenish colour in the whole head	**2**
		All shrimp, very dark, yellowish, greenish, dark colour of the head extends to the tail part.	**3**
	Colour (abdomen and tail)	Rather pale pink, red, bright/clear colour	**0**
		Pale pink, more matt	**1**
		Brownish, Yellowish	**2**
		Brown, yellow, green-, greyish discolouration	**3**
	Odour	Fresh, seaweady	**0**
		Faint odour, reminds of tar	**1**
		Faint ammonia odour, fishy	**2**
		Obvious ammonia odour, putrid	**3**
Roe	**Roe colour**	Copper green	**0**
		Discoloured, faded, yellowish	**1**
		Dark, yellowish	**2**
Quality Index			**0-11**



Day 0: Pink, bright/clear appearance; green roe
Day 1: Pale pink abdomen and tail; slight brownish/yellowish discolourations on the head; green roe
Day 3: Brown, yellowish, greenish colour in all heads; brownish/yellowish abdomen and tail; discoloured/faded roe
Day 7: Dark, yellow, green, greyish discolourations extending from the head to the abdomen and tail; dark, yellow roe

Figure 14. Changes occurring in thawed fjord shrimp at different storage days in ice.

4.2 Shelf-life study of thawed fjord shrimp stored in ice

4.2.1 Sensory evaluation

The developed QIM scheme for thawed fjord shrimp was used to evaluate the samples during sensory sessions. Evaluation of the samples on storage days 0, 1,3,5,6 and 8 was completed in two sessions. Samples at storage day 0 were bright pink, had a copper-green roe, fresh sea weedy odour and a slight brownish colour on small/no part of the head. As the storage time increased from 5 to 8 d, the heads of the shrimp became very dark with greenish and yellowish discolorations extending to the abdomen and tail, the odour became putrid, and the roe faded, turned dark, and yellowish.

There was a linear increase in the QI scores with storage in ice (day 0-8) for all the quality parameters evaluated (Figure 15). For each of the quality parameters evaluated, the scores were the lowest for samples on storage day 0 and the highest for samples on storage day 9 (Table 3). The roe colour and colour of the abdomen/tail for samples on storage day 6 were lower than those of day 5 samples. All the parameters had a mean score of 0-0.5 for samples stored at day 0, and a mean score of 2.4-2.8 for samples at storage day 8, except for the roe colour which had a mean score of 1.5 because of its 2-point grading scale. Samples of storage day 8 had the highest mean score given for "Dark in the head" and the lowest score for roe colour.

Table 3. Average scores with standard deviation of each quality parameter evaluated per storage day in ice.

Storage Days	**Dark in the head**	**Colour (abdomen/tail)**	**Odour**	**Roe colour**
0	0.2±0.1	0.4±0.6	0.3±0.4	0.3±0.6
1	0.5±0.7	0.4±0.7	0.6±0.5	0.5±0.5
3	1.7±0.6	1.4±0.4	1.4±0.4	1.2±0.5
5	2.1±0.5	1.8±0.6	1.5±0.6	1.4±0.4
6	2.6±0.5	1.6±0.3	2.0±0.3	1.4±0.5
8	2.8±0.3	2.5±0.5	2.4±0.4	1.5±0.4

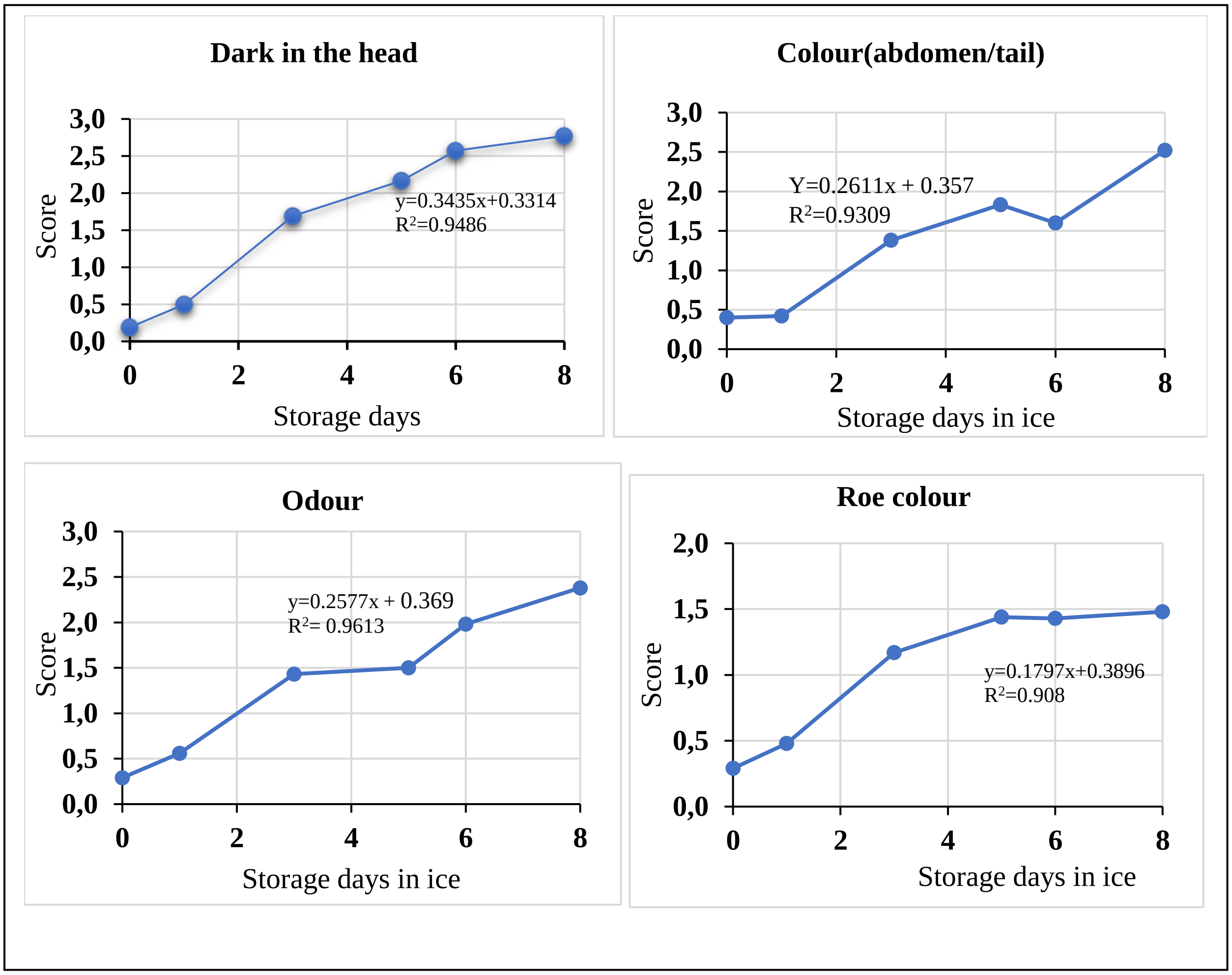


Figure 15. Average score for each quality parameter evaluated with QIM scheme for thawed fjord shrimp stored on ice.

Nine panellists evaluated the samples in two sessions of the shelf-life studies. Five trained and two untrained panellists participated in the first sensory evaluation session on storage days 0, 3, and 6, respectively. During the second evaluation session for samples on storage days 1, 5, and 8, panellist 2 from the first sensory evaluation was absent, whereas two new untrained panellists joined (panellist 8 and 9). There were slight variations in the mean QI scores provided by the panellists (Figure 16). Panellist 7 gave very high scores for samples from the lowest storage days, whereas panellist 1 gave the highest score for day 5 samples. Panellists 5 and 7 both scored day 5 samples higher than day 6 samples. Panellist 6 scored day 0 higher than days 1 and 3, which was higher than day 5. Panellists 3 and 4 performed the best during the evaluation sessions. Five out of the seven panellists gave their lowest scores on storage day 0, whereas seven out of the eight panellists gave their highest scores on storage day 8 (Appendix 1). This means that the panellists agreed more when evaluating samples on storage day 8.

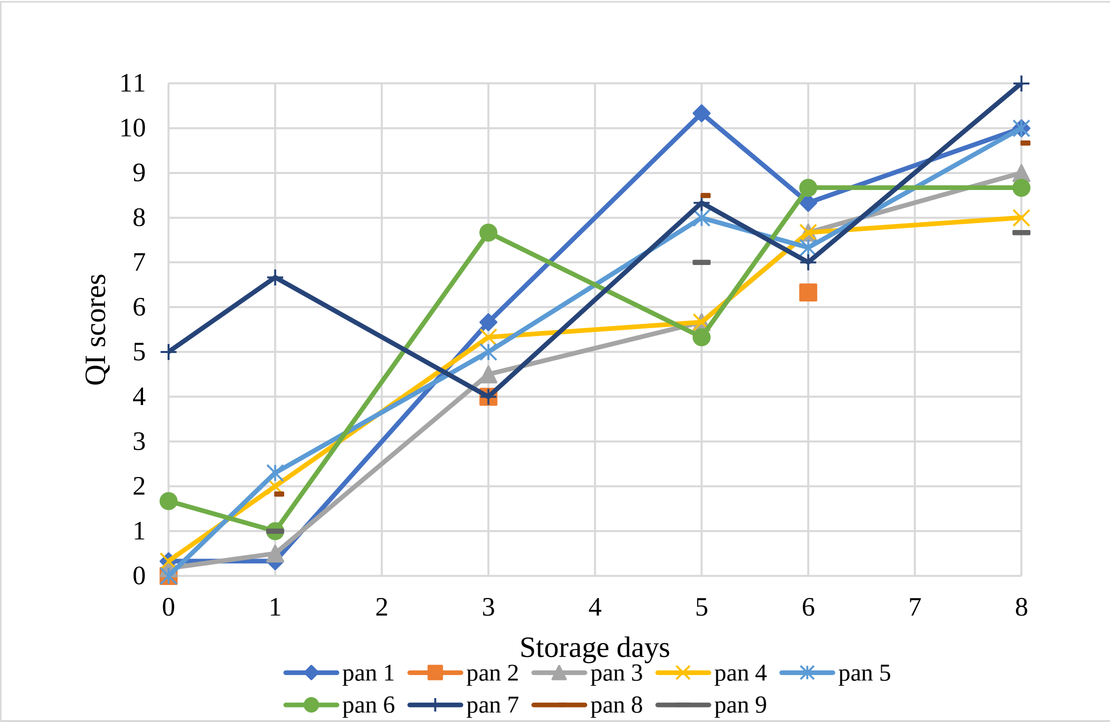


Figure 16. Quality index (QI) scores per panellist against storage days in ice.

The average QI scores and standard deviations for whole shrimp on storage days 0, 1,3, 5, 6, and 8 are presented in Table 4. This was calculated as the mean score of all four quality parameters evaluated by the panellists, except panellist 7, due to deviation from the scores given by the panellist. As the number of storage days of the samples in ice increased, the mean QI score also increased. A linear correlation (R^2=0.9487) was found between QI and storage days in ice (Figure 17). At the end of the storage period, the Quality Index value was 9.0, which is close to the maximum demerit point of 11.

Table 4. Average QI scores for each sample per storage day.

Storage Days	Mean QI
0	0.4±0.6
1	1.3±0.8
3	5.4±1.3
5	7.2±1.8
6	7.7±0.8
8	9.0±0.9

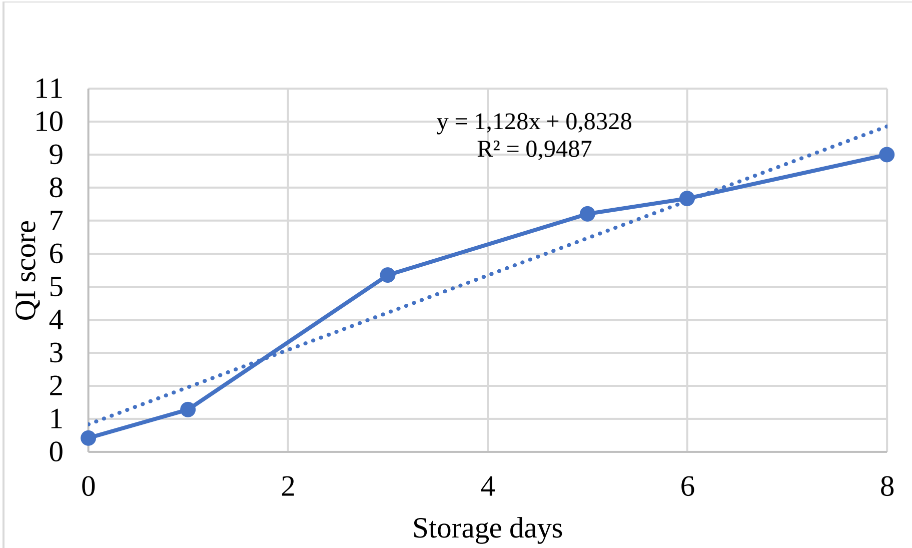


Figure 17. Quality index (QI) score of thawed fjord shrimp based on evaluation of 8 trained sensory panellists (without panellist 7), storage days in ice.

During the evaluation sessions, samples from each storage day were analysed in triplicate. Good consistency was observed among the triplicate samples from all storage days (Table 5). The lowest QI score of 0.79 was given for a replicate of storage day 0 and the highest 9.69 for a replicate of storage day 8. A Regression correlation of R^2 = 0.9445 was found between the replicates and storage days on ice (Figure 18).

Table 5. Average QI scores of each sample replicated with storage days.

Storage days	Average QI score/Standard deviation		
	Replicate 1	**Replicate 2**	**Replicate 3**
0	0.86±1.21	0.79±1.87	1.57±2.94
1	0.94±1.52	2.35±2.12	2.69±2.80
3	5.64±1.93	5.08±0.92	5.50±1.50
5	6.81±1.73	7.81±2.14	7.44±1.99
6	7.71±1.11	7.36±1.11	7.64±0.63
8	9.25±1.28	8.69±1.53	9.69±1.03

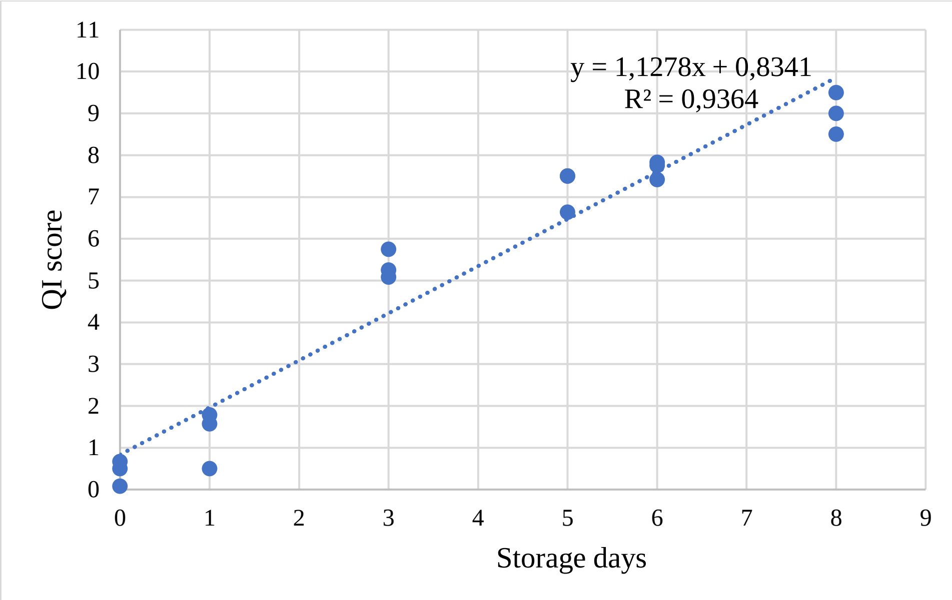


Figure 18. Distribution of QI scores of each sample replicates with storage days in ice.

4.2.2 Microbial Analysis

Microbial analysis of the samples showed an increase in Total Viable Counts (TVC) and hydrogen sulphide-producing spoilage bacteria (H_2S) with storage time (Figure 19). TVC increased from log 4.89 Cfu/g on storage day 0 to log 8 Cfu/g on storage day 8, which is higher than the recommended limit of Log 6 Cfu/g (ICMSF, 1978), while H_2S bacteria increased from log 3.43 Cfu/g on storage day 0 to log 6.30 Cfu/g on storage day 8 (Appendix 2). A high proportion of the total viable count was due to spoilage bacteria.

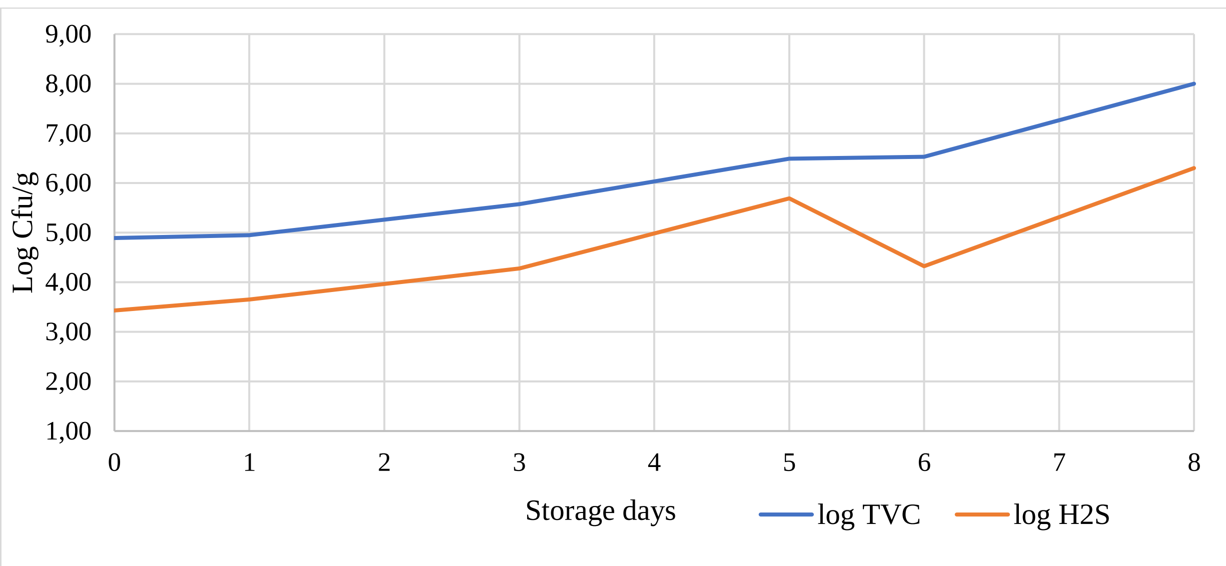


Figure 19. Total viable counts and counts of H_2S-producing bacteria against storage days.

4.2.3 Chemical analysis

TVB-N generally increased throughout the storage period. The TVB-N content in samples on storage day 0 was found to be 13.50 mg N/100 g, which increased to 43.10 mg N/100 g at the end of the storage period (Figure 20). There was a slight fluctuation in TVBN values between storage days 1 and 6. There is no estimated TVB-N content limit for shrimps (Dordevic & Buchtova, 2016)). However, the European Union regulations (EC, 2008) state that the limits allowed for unprocessed fishery products are 25-35 mg N/100 g. Samples of storage days 5 and 6 with values of 29.40 mg N and 28.70 mg N respectively were above the lower recommended limit while samples of storage days 1, 3, and 8 with values of 39.30 mgn/100 g, 36.40 mg N/100g and 43.10 mg N/100g respectively were above the upper recommended limit.

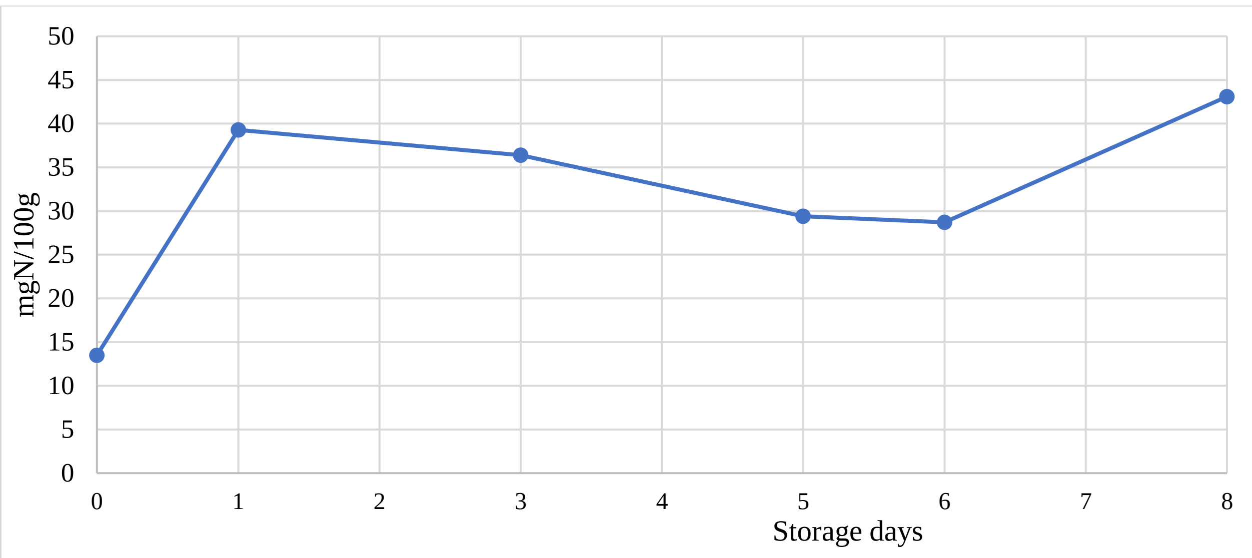


Figure 20. TVB-N values of thawed fjord shrimp stored on ice.

4.3 Development of Preliminary QIM scheme for brown (*Parapenaeopsis atlantica)*, white (*Penaeus notialis*) and tiger (*Penaeus monodon*) and shrimp species

The shrimp photos sent to Matis from Nigeria were carefully pre-observed for visual changes occurring during storage on ice for nine days. Visible colour and odour changes were observed during storage in ice. The brown shrimp appearance changed from bright brown/greyish body on storage day 0, to black spots (head) and slight discoloration of the body on storage day 2, to dark, brownish/yellowish/black discoloration on the heads, legs, and some parts of the abdomen on storage day 4, and finally to a completely dark, black/yellow discoloration on the whole shrimp on the 9th day of storage (Figure 21).

The appearance of white shrimp changed from bright grey/brown body on storage day 0, to dull, slightly discoloured body with black spots on some heads on storage day 2, to yellowish and black spot discoloration on the heads and parts of the abdomen/tail on storage day 4, and finally to black spots (on all heads), very back legs, and pale/yellowish body on the last day of storage (Figure 22).

The tiger shrimp appearance changed from a bright grey body and reddish legs on storage day 0, to a dull body on storage day 2, to the heads becoming dark on storage day 4, and finally the whole shrimp appeared dark with reddish heads on storage day 9 (Figure 23). A maximum of 9, 12, and 11 Quality Index points were recorded for the brown, white, and tiger shrimp species, respectively.

The changes in the sensory attributes observed were used to develop a preliminary QIM scheme for each of the three different species (Tables 6, 7, and 8), which will be used for training panellists and finalizing the schemes in Nigeria.

Table 6. *Preliminary Quality Index Method (QIM) scheme for thawed brown shrimp.*

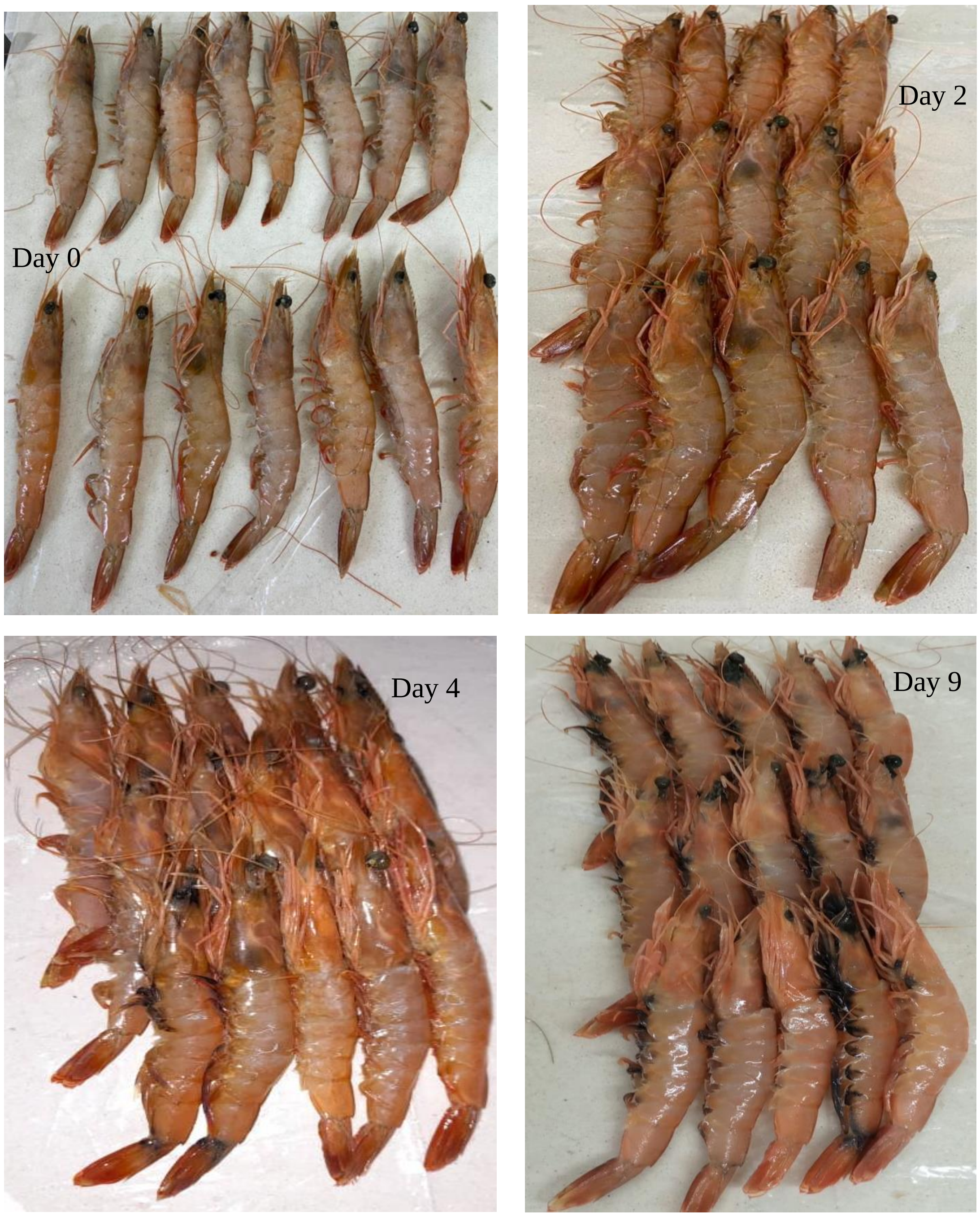
Quality parameter		**Description**	**Score**
Whole Shrimp	**Dark in the head**	None	**0**
		Some (25%)	**1**
		Many (50-75%)	**2**
		All (75-100%)	**3**
	Colour	Light brown, grey, bright, and transparent body, bright yellow stripe on tail	**0**
		Dull, slight discoloration on the body, dark tail with dull yellow stripes	**1**
		Dark brownish, blackish, yellowish discolorations	**2**
		Almost black, yellowish discolorations	**3**
	Odour	Fresh, seaweady	**0**
		Faint odour, reminds of tar	**1**
		faint ammonia odour, fishy, sour	**2**
		Obvious ammonia odour, sour, putrid	**3**
Quality Index			**0-9**

Day 0: Brown/greyish bright/clear body
Day 2: Appearance of black spots on the head, slight discoloration on the body
Day 4: Dark brownish appearance, yellowish, and black discolorations on heads and legs
Day 9: All heads turned black, yellowish, and black discolorations on whole shrimp

Figure 21. Changes occurring in thawed brown shrimp at different storage days in ice.

Table 7. Preliminary Quality Index Method (QIM) scheme for thawed white shrimp.

Quality parameter		Description	Score
Whole Shrimp	**Black spots in the head**	None	**0**
		Some (25%)	**1**
		Many (50-75%)	**2**
		All (75-100%)	**3**
	Colour	Mustard brown, appears bright and transparent	**0**
		Almond brown, appears dull, slight discoloration around the appendages	**1**
		Appears dull, blackish, yellowish discolorations	**2**
		pale, yellow, greyish discoloration, black appendages	**3**
	Black spots in the tail	None/slight (0-25%)	**0**
		Some (25-50%)	**1**
		Many/large areas of tail part (50-75%)	**2**
		All (75-100%)	**3**
	Odour	Fresh, seaweady	**0**
		Faint odour, reminds of tar	**1**
		Faint ammonia odour, fishy, sour	**2**
		Obvious ammonia odour, sour, putrid	**3**
Quality Index			**0-12**

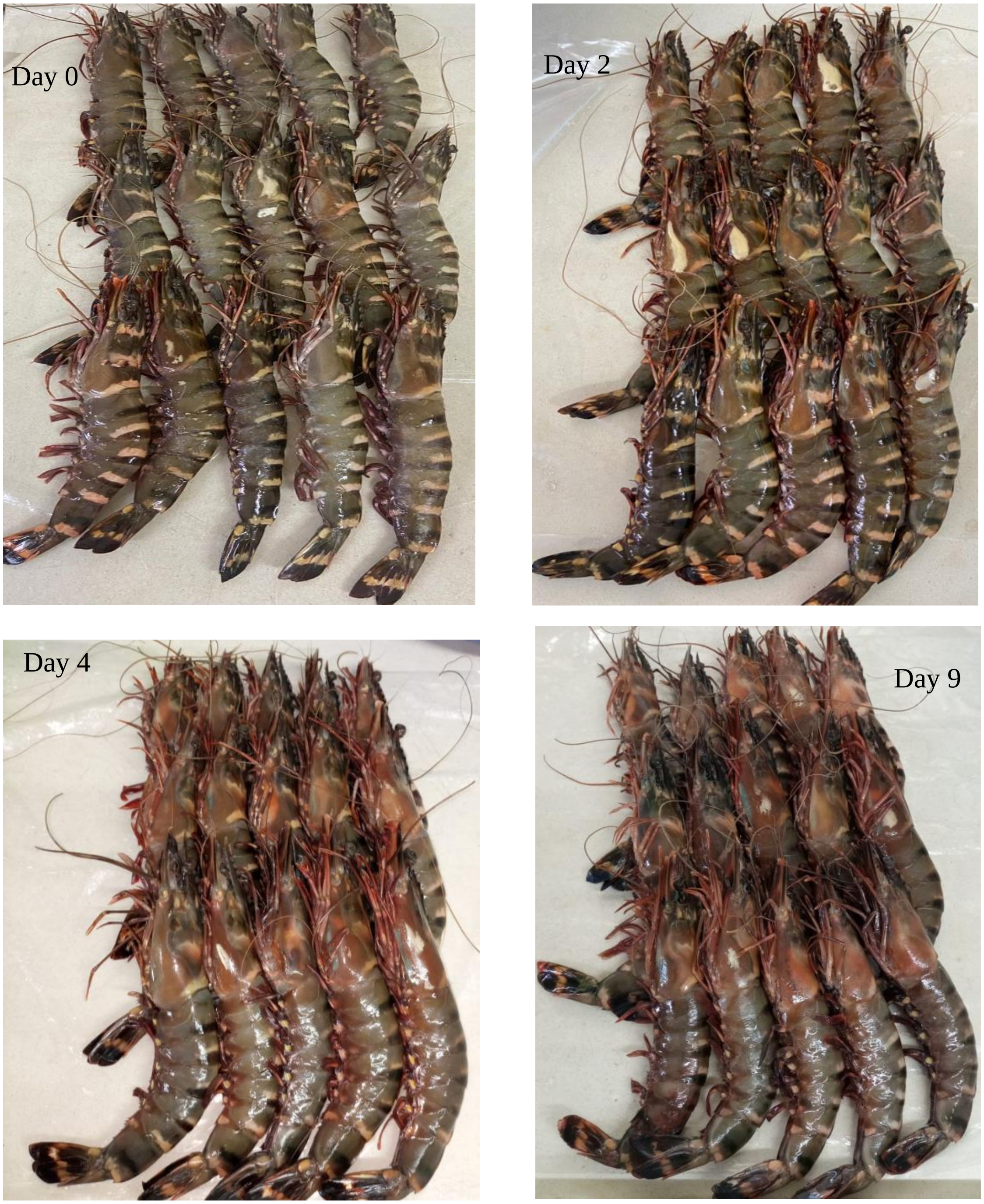


Day 0: Brown, grey bright/clear body
Day 2: Slight discolorations, dull appearance, black spots on some heads
Day 4: Yellowish and black spots discolorations on the heads, parts of the abdomen and tail
Day 9: Black spots on all heads, shrimp body pale/yellow, all legs very black

Figure 22. Changes occurring in thawed white shrimp at different storage days in ice.

Table 8. Quality Index Method (QIM) scheme for thawed tiger shrimp.

Quality parameter		Description	Score
Whole Shrimp	**Dark/Red in the head**	None	**0**
		Some (25%)	**1**
		Many (50-75%)	**2**
		All (75-100%)	**3**
	Colour	Brightly coloured grey with white and black band	**0**
		Dull, grey colour with pinkish/ milk colour bands	**1**
		Dull, brownish discolorations, pinkish bands	**2**
		Very dull, yellowish, brownish/reddish discolorations	**3**
	Colour (Legs)	Greyish and transparent	**0**
		Slight Reddish discoloration	**1**
		Reddish/Blackish	**2**
	Odour	Fresh, seaweady	**0**
		Faint odour, reminds of tar	**1**
		faint ammonia odour, fishy, sour	**2**
		Obvious ammonia odour, sour, putrid	**3**
Quality Index			**0-11**



Day 0: Grey with white and black bands, bright/clear body, reddish legs
Day 2: Dull appearance
Day 4: Dark shrimp heads with brown discolorations
Day 9: whole shrimp dark with all heads reddish

Figure 23. Changes occurring in thawed tiger shrimp at different storage days in ice.

5 DISCUSSION

5.1 Development of QIM scheme for thawed fjord shrimp

Pre-observation of thawed fjord shrimp was carried out to modify the QIM scheme for whole fresh and unfrozen fjord shrimp (Martinsdottir, Sveinsdottir, Luten, Schelvis, & Hyldig, 2001). The reason for the pre-observation and modification was to observe changes that are likely to occur during freezing/thawing, which could influence the spoilage pattern. Freshly thawed shrimp were observed to have slightly dark heads and pale bodies compared to fresh unfrozen shrimp (Lei Jin, Guofang, Peipei, Jie, & Xiaojun, 2018) reported similar changes observed in thawed marine trawling shrimp stored at deep frozen temperatures which was due to the freezing.

During training, the description for the parameter "dark in the head" was modified to better suit the changes in appearance of the head with an increase in storage days. The darker greenish-yellowish colour is due to the production of diffusible pigments by microbiological activities (Gram & Huss, 1996). The body part (abdomen and tail) was added to the "colour" parameter for better description. The sour descriptor was removed from the odour quality parameter because none of the samples were perceived to be sour throughout the storage period. The final QIM scheme for thawed fjord shrimp included four quality parameters and 11 demerit points.

5.2 Shelf-life studies

5.2.1 Sensory evaluation

The sensory changes observed in whole shrimp from the first to the last storage days in ice indicated quality deterioration of the shrimp. According to Du, Chai, Guo, & Lu (2015), shrimp change colour and produce an unpleasant odour as its quality changes. This result showed that all four quality parameters (dark in the head, colour of the abdomen/tail, odour, and roe colour) evaluated were useful in assessing the quality deterioration of the shrimp, as they all showed a clear linear relationship with storage time (Figure 15). The quality parameter "dark in the head" was scored the highest from storage days 3 to 8, which means that the quality of the head was lost faster compared to other parts of the shrimp (Table 3). According to Banrie (2013), "the enzymes that degrade prawns are mainly digestive enzymes located in the prawn's head". The average QI calculated for the whole shrimp from the scores (sum of all attributes) of all the panellists except one (panellist 7) showed a high linear correlation (R^2=0.9487) with storage days in ice, which is in line with the findings of Ainaz, (2019) who also found a high linear correlation of R^2 = 0.983 between the total QI and storage days in ice of *Metapenaeus affinis*. This showed that the quality of whole shrimp decreased with an increase in storage time. On storage day 5, shrimp became unfit for human consumption. This result showed that the shelf life of thawed fjord shrimp is shorter than that of whole fresh unfrozen fjord shrimp, with a shelf life of 6 days during storage in ice (Martinsdottir, Sveinsdottir, Luten, Schelvis, & Hyldig, 2001). Depok (2014) also reported that the shelf life of ice and shrimp ratio 1:1 and ice to shrimp ratio 1:3 samples was 5 days. A good consistency was observed among each sample replicates, which demonstrates the importance of having a minimum of three replicate for samples of each storage day (Bonilla, Sveinsdottir, & Martinsdottir, 2007). Differences observed in the scores given by each panellist were expected, as this was the first time a QIM scheme for thawed fjord shrimp was used, apart from the training sessions. The panellists who had wide variations in their scores and performed poorly were untrained panellists, thus emphasizing the need for proper training if they were to perform very well during sensory evaluations.

5.2.2 Microbial Discussion

The microbial result showed a continuous increase in TVC and H_2S bacteria from 4.89-8.0 Log Cfu/g and 3.43 - 6.3 Log Cfu/g respectively as the storage days progresses (Figure 19). This result is in line with the findings of Ainaz (2019), who reported a continuous increase in total mesophilic bacteria (TMC) and psychrotrophic total counts (PTC) in *Metapeneaus affinis* shrimp species stored on ice as storage time increased. Zeng, Thorarinsdottir, & Olafsdottir (2005), also reported a steady increase in TVC of *Pandalus borealis* during storage in ice. Spoilage of chilled seafood is mostly caused by gram-negative psychrotrophic bacteria such as *Shewannella* and *Pseudomonas* species, which give off odours in fish products (ICMSF, 2011). This study showed that, throughout the storage period, a high proportion of the total viable count was spoilage bacteria (Appendix 2). At storage day 5, the TVC value of 6.49 Log Cfu/g exceeded the recommended acceptable limit of log 6 Cfu/g in shrimp (ICMSF, 1978). The microbial results of this study are in good agreement with the sensory results.

5.2.3 Chemical analysis

The result of the measurement of TVB-N value increased from 13.50 mg N/100 to 43.10 mg N/100g over the storage period (Figure 20). Ainaz, (2019) reported a similar increase of TVB-N from 11.89 mg N/100g to 39.59 mg N/100g during a 12-day storage period of *Metapenaeus affinis* in ice. According to Ainaz (2019), the increase in TVB-N compounds may be related to protein and nonprotein degradation by bacteria in shrimp. High levels of TVB-N were measured in samples from storage day 1 to 8 (Appendix 3), which was similar to the findings of Zeng, Thorarinsdottir, & Olafsdottir (2005) who also reported a high TVB-N value of 33.5 mgN/100 g for whole northern shrimp at the beginning of storage in ice. According to Zeng, Thorarinsdottir, & Olafsdottir (2005), the use of too little ice could minimize the washout of TVB-N, resulting in high TVB-N values in the samples. A low initial TVB-N of 13.50 mg N/100 g for storage day 0 indicates superior freshness (Dai, Cheng, Sun, Zhu, & Pu, 2016). The TVB-N values for samples on storage days 5 and 6 were above the lower limits, while storage days 1,3 and 8 were above the higher limits of 25-35 mg N/100 g recommended by the European Union regulations (EC, 2008) for all unprocessed fishery products. In general, TVB-N levels are used to determine the freshness of shrimp, but the results from this study further confirm that TVB-N content alone cannot be considered a freshness indicator but must always be corroborated by sensory tests (Condurso et al., 2016).

5.3 Development of Preliminary QIM scheme for thawed white (*Penaeus notialis*), tiger (*Penaeus monodon*) and brown (*Parapenaeopsis atlantica)* shrimp species

As the storage period progressed, visual sensory changes were observed in shrimp photographs from Nigeria. According to Mu, Fang, Mao, Gao, & Chen, 2012; Du, Chai, Guo, & Lu (2015), the formation of black spots, yellowish discolorations, and fishy/putrid odour observed during storage in ice indicated quality deterioration of the shrimp. All shrimp had no visible roe and as such "roe colour" was not part of the quality parameters described. The preliminary QIM scheme developed for the tiger shrimp had the same number of QI demerit points with the QIM for fresh unfrozen whole fjord shrimp (Martinsdottir, Sveinsdottir, Luten, Schelvis, & Hyldig, 2001), unlike the brown shrimp which had a lower QI demerit point, and the white a higher QI demerit point. The schemes for white and tiger shrimp species had higher QI demerit points than the brown shrimp, because separate descriptions were given for visible colour changes appearing on the tail and legs of the white and brown shrimp, respectively.

6 CONCLUSION AND RECOMMENDATION

This study was conducted because of the importance and need for a freshness grading scheme for commercially important shrimp species in Nigeria and an established training protocol for use within the quality assurance unit of the Federal Department of Fisheries, Nigeria.

Preliminary QIM schemes were developed for brown (*Parapenaeopsis atlantica*), white (*Peneaus notialis*), and tiger (*Penaeus monodon*) shrimp species based on photos from Nigeria. The QIM scheme developed for white and tiger shrimp included 4 quality parameters with a total of 11 and 12 demerit points respectively, and the QIM scheme developed for brown shrimp included 3 quality parameters with a total of 9 maximum demerit points.

A QIM scheme was developed for thawed fjord shrimp in a shelf-life study, which revealed a linear relationship with a high correlation between the quality index (QI) and storage days in ice. The growth of bacteria (TVC and H_2S) also increased with the storage time. The measurement of TVB-N content did not show a linear correlation with storage duration; hence, TVB-N is not suggested as a reliable quality parameter for evaluating the freshness of thawed fjord shrimp in ice. This study showed that sensory evaluation and microbial counts are good parameters for determining the quality of freshness and thus estimating the shelf life of thawed fjord shrimp stored in ice. Sensory results at storage day 5 showed that most of the shrimp had brown, yellowish and greenish colour on the head, faint ammonia/fishy odour with discoloured/faded roe while the TVC count of Log 6.49 Cfu/g was above the recommended limit of Log 6 Cfu/g. Based on the sensory and microbial results, the point of rejection of the thawed fjord shrimp sample stored on ice was five days. It is therefore recommended that after thawing, shrimp be stored in ice at cold temperatures to maintain its freshness and prolong its shelf life. QIM has been shown to be a leading practical tool that can be developed to evaluate shrimp freshness in the fish sector and fish inspection services in Nigeria and other African countries.

Based on the knowledge gained during training sessions in shelf-life studies, standard protocols/guidelines for the selection and training of assessors and plans for implementation and finalization of the QIM schemes in Nigeria were established (Appendix 4). QIM is well suited to teaching inexperienced people how to evaluate fish and shellfish. It is also suitable for teaching panellists and monitoring their performances. Using the standard protocols and guidelines in the training and re-training of assessors in quality assurance, fish inspection services are recommended for assessors to perform very well during sensory evaluation. Finalization and implementation of the QIM scheme in Nigeria would be useful for shrimp quality control and strengthening quality assurance fish inspection services. It is also recommended that QIM schemes be developed for other commercially important shrimp and fish species based on different quality attributes, spoilage indicators, and quality standards of each species.

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APPENDICES

Appendix 1. Average QI scores per panellist

Storage Days	Pan 1	Pan 2	Pan 3	Pan 4	Pan 5	Pan 6	Pan 7	Pan 8	Pan 9
0	0.33	0	0.17	0.33	0	1.67	5	1.83	
1	0.33		0.5	2	2.3	1	6.67		1
3	5.67	4	4.5	5.33	5	7.67	4	8.5	
5	10.33		5.67	5.67	8	5.33	8.33		7
6	8.33	6.33	7.67	7.67	7.33	8.67	7	9.67	
8	10		9	8	10	8.67	11		7.67

Appendix 2. Log values of TVC and H2S with storage days on ice

Storage days	Log TVC	Log H_2S
0	4.89	3.43
1	4.95	3.65
3	5.57	4.32
5	6.49	5.69
6	6.53	4.32
8	8.00	6.30

Appendix 3. Log values of TVB-N with storage days on ice

Storage days	TVB-N
0	13.50
1	39.30
3	36.40
5	29.40
6	28.70
8	43.10

Appendix 4. Training protocol/guideline

Scope and purpose of the guidelines/training protocol

The training protocol is required to guide fisheries regulatory agencies, which in Nigeria are the Federal Department of Fisheries and Aquaculture, and quality control managers of commercial seafood companies on knowledge of procedures used in sensory evaluation of shrimp freshness, to facilitate and enhance shrimp quality control. To avoid errors in the sensory evaluation, it is necessary to follow well-defined guidelines and standards.

For the successful implementation and daily use of sensory evaluation in quality control, it is important that the following is in order:

- ✓ Approval and acceptance of the scheme by the head of the quality assurance section or the quality control manager
- ✓ Storage and evaluation facilities
- ✓ Sampling plan for daily quality control including number of samples, method of collection and evaluation procedures
- ✓ Recruitment of assessors, selection, and training
- ✓ Training for the use of QIM schemes and finalization of QIM schemes for landed species.

The following sub-headings describe in more details the above listed steps

Required Facilities for Sensory Evaluation

Facilities required for sensory evaluation should be located within seafood companies and must be easily accessible.

Preparation area
This area is used to prepare shrimp samples for sensory evaluation. It is also used for the handling and storage of fishery products. To avoid bias judgment, this area should be separate from the actual testing or evaluation area so that the assessors do not interfere with the sample preparations.

Evaluation area
This area is used to evaluate fishery products. Sample preparation and chemical and microbial analyses should not be performed in this area. The area should be well ventilated (artificial air conditioners can be provided), spacious, free of foreign odours, far away from noise and disturbances, and the whole room should have adequate bright lightning. The colour of the walls should be even, preferably off-white.

Sensory Panel leader

Having a well-trained leader who coordinates all sensory evaluation activities is crucial. This individual, known as the panel leader, should have an academic background in fisheries quality control or related disciplines. They must be well trained in performing sensory evaluations of shrimp and other seafood. The panel leader is usually in charge of the selection and training of panelists in the basic and descriptive tests. The panel leader is also charged with the responsibility of organizing and implementing training programs, monitoring and assessing panellists' performance, ensuring adequate supplies of training samples, and ensuring the maintenance and updating of all relevant documentations and motivation of the panellists. The panel leader also observes that sensory evaluation is conducted in a professional manner. He/she gives reports or review for quality control management decisions (ISO 13300-1, 2006)

Selection of assessors for training

Assessors trained for sensory evaluation are selected based on the ISO 8586 (2012) standards. To qualify for selection, the assessor must

1. be able to perceive odours, so that the odours of decomposition will be perceived.
2. have normal colour vision and can detect abnormalities in the appearance of fishery products.
3. be able to rely on sensory perceptions and appropriately report them.
4. be able to learn terminology for new or unfamiliar perceptions (odours, tastes, appearance, and textures).
5. be able to define sensory stimuli and relate them to the underlying cause of the product.

Screening for perception of odour

This should be based on ISO 8586 (2012) standard guidelines. About 5-10 olfactory stimuli, preferably related to the product or products intended to be evaluated, should be prepared. The stimuli should contain samples that are easily recognized and others that are less common. The intensity of these stimuli should be well above the recognized threshold but not significantly above the levels encountered during the sensory evaluation of the product itself. Stimuli such as vinegar (acetic acid), seaweed, tar, and/or TMA-trimethylamine are recommended for the evaluation of raw shrimp. Samples for odour perception should be hidden, for example, under cotton in an odourless flask/container. The lid of the containers/flask should be properly sealed to avoid odour evaporation, and all samples should be coded. Participants should be given an evaluation form and asked to identify or describe the odour they perceive from each stimulus to the best of their ability.

Screening for perception of colour

The aim is for the assessor to identify different shades of colour, in relation to the colour changes that may occur when spoilage of seafood sets in. This is based on ISO 11037 (2011) standard guidelines. Samples of different colour related to the product to be evaluated should be presented to the participants for identification. For shrimp, samples with colours such as green, grey, yellow, and different shades of brown should be used. Participants should be given an evaluation form and asked to identify the colour they see.

The evaluation form should contain the following information.

Date: ……….. **Your name**: ………………………………

Sample no	**Odour/Colour Sample description**
_____	____________________________________
_____	____________________________________
_____	____________________________________

Potential panellists should be graded according to performance on a scale: 3 points for a correct identification or a description of the most frequent association, 2 points for a description in general terms, 1 point for an identification or description of an appropriate association following discussion, and 0 points for no response or a totally wrong response. A satisfactory level for this task can be specified only in relation to the materials used. Each participant must receive at least 65% of the maximum score to be fit as a panellist.

General/Basic rules for sensory evaluation

- Eating, drinking, and smoking should not be permitted in the test area.
- It is necessary to rest between the samples and breath fresh air when evaluating the sample odour.
- Smoking 30-60 minutes before the sensory evaluation is highly prohibited.
- All samples for sensory evaluation should be coded with 2–3-digit numbers before being presented to the panellist.
- 10-11am and 2-3pm are considered the best times for sensory testing.
- Panellists should understand the importance of sensory evaluation in quality control.
- The preparation area should be close to the evaluation and testing area.
- People with health challenges should not be allowed to participate in sensory evaluations.
- Interactions among the panellists during sensory testing are prohibited.

Sample collection/ handling/preparation

Different grades of shrimp should be randomly selected from each batch (CAC/GL 31, 1999). During the collection of frozen samples for training and evaluation, the inspectors should ensure that the samples are handled with care so that the sensory properties are not affected. The samples must be properly packed and, where necessary, under temperature control before being taken to the sensory laboratory (CAC/GL 31, 1999). Frozen samples must be thawed and evaluated immediately in the laboratory; otherwise, they should be stored under the appropriate conditions. However, fresh and chilled products must be evaluated on the day that they are received. Samples stored in ice should be well wrapped or packed to avoid desiccation or drying out. Prior to evaluation, frozen samples must be thawed at ambient temperature by spreading them on work surfaces in the preparation area.

Training procedure

Once the panellists have been selected, the preparation of the samples to be evaluated should be performed. The panel leader starts by describing the procedures of the sensory evaluation, what is expected of the panellists, and so on. The panel leader should carefully explain the QIM schemes intended for use in freshness evaluation and should also explain the importance of breathing deeply and resting between samples during odour evaluation. Panellists should be encouraged not to let their personal judgement interfere with the evaluation process. Three to four samples of shrimp of different freshness with known storage times in ice should be used. During the first training sessions, the samples should not be coded but should be coded during the second and third training sessions to enable the evaluation of the panellist's performance. During the training, discussions are allowed between the panel leader and panellists. Two to three training sessions are recommended to improve panelists' performance. Revision/retraining should be done 1-2 times per year.

Finalization of the QIM scheme for thawed brown, white and tiger shrimp species

The preliminary QIM schemes developed for white (*Peneaus notialis*), brown (*parapenaeopsis atlantica*), and tiger (*Penaeus monodon*) shrimp species within the GRO-FTP at Matis will be used to train assessors (quality assurance inspectors) from the Quality Control Division of the Federal Department of Fisheries in Nigeria. On arrival at the sensory laboratory, the frozen shrimp samples shall be thawed for approximately 3 hours at ambient temperature by spreading them out on a clean surface in the preparation area for defrosting. The thawed samples shall be stored at a temperature of 0-4^{0}C in a refrigerator or on ice for nine days. Samples shall be taken on storage days 0, 2, 4, 7, and 9 to obtain different levels of freshness. 10-15 shrimp from each level of freshness shall be placed on the sensory evaluation table under bright light.

During the first training session, the panel leader shall explain to the panellists the different quality parameters on the preliminary QIM scheme and describe the sensory changes to look out for amongst different samples on different storage days. A copy of the scheme shall be given to each panellist, and they should consider how to assign scores for each quality parameter evaluated. The panellists shall be allowed to interact between themselves and with the panel leader regarding their suggestions and opinions. Immediately after this training session, the samples shall be kept under the same storage conditions prior to the evaluation. A 30-to 40-minute break shall be taken to make the necessary changes and adjustments to the preliminary scheme.

The modified preliminary QIM scheme shall be used during the second training session to evaluate the samples as before, but this time, coded. Further adjustments and changes should be made to the descriptors or quality parameters, where appropriate. At the end of the two training sessions, the final QIM scheme shall be developed. This process shall be conducted separately for all three species. The panellists should be motivated and appreciated with words of encouragement and the provision of refreshments.

Monitoring of Assessors

The panel leader shall monitor the performance of the panellists during the sensory training sessions according to (ISO 6658, 2017). Sensory data from the training sessions shall be analysed and interpreted by the panel leader, sensory staff, or project leaders. Statistical analysis (averages, Analysis of Variance, and t-tests) shall be conducted using the scores given by the panellists during the training session to determine the performance of each panellist. Panellists who performed poorly shall be booked for additional training sessions.

Adoption/Implementation of the QIM schemes

The QIM schemes developed for all three species should be adopted for grading shrimp freshness by the quality assurance inspection service of the Federal Department of Fisheries and quality control personnel of the shrimp industry. The FDFA-trained inspectors shall in-turn train the quality control team in these shrimp companies.

The importance of a QIM scheme and its contribution to quality control management should be explained to the head of the quality assurance unit of the FDFA. It is expected that the idea will be bought without hesitation, and the usage of the QIM schemes will be implemented immediately after its finalization. The entire process of selling the idea to the unit head, selection and training of panellists, finalization, and implementation of the QIM scheme should take to 2-3 months for completion.