Effects of Storage on Product Quality of Bivalves (oysters, scallops, and clams)

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Executive Summary

The purpose of this thesis is to investigate the effects of the different storage conditions concerning quality of oysters *Ostrea edulis*, scallops *Pecten maximus* and clams *Arctica islandica*. Quality of bivalves is a very broad concept dependent of several factors such as the preferences of the consumer, the conditions which the animals have grown, harvest methods as well as the level of stress post-harvest. The organoleptic characteristics, such as the appearance and smell, are very important in the perception of quality by the consumer, and these factors might be the most decisive for the acceptance of the food product. The bivalves which are harvested during different seasons might have different quality. Seasonality can have influence on the sensorial characteristics since there are biochemical changes in the body of these molluscs throughout the year, due to the feed and the reproduction stages. Moreover, the bivalves which are subject to high levels of stress conditions will be of lower quality. The stress, whether due to environmental factors (food availability, salinity, temperature and water quality) or due to the harvest and post-harvest (fishing methods, storage conditions), will influence survival during storage and in therefore, influence the quality of the final product.

In this study, wild bivalves were stored alive in polypropylene boxes at different temperatures (3, 6, and 9 ºC) during different days (3, 6, 9 and 12 days) and the quality was verified through a sensory evaluation, weight assessment and survival. The evaluation of quality was carried out with two different sizes of oysters, two different sizes of scallops and one size of clams. In addition, was verified the relationship between the quality, and the season variability.

The results showed that the variability of the oysters' quality between seasons, temperature and time of storage were not a significant ($p > 0.05$). The survival rate was high during the storage (6 deaths in 168 oysters) and the influence of the storage conditions was not significant. Regarding to scallops, it was concluded that with the increase of both temperature and time of storage, quality decreases. Scallops can be stored until 7 days at 3 ºC without loose quality and only until 3 days at 6 and 9 ºC. Once, after the day 3 at 6 and 9 ºC, all scallops died can be concluded that the survival of the scallops and the storage conditions were related. Moreover, the season which the scallops were harvested was significant in the final quality of the product, being August the best season for harvesting. Regarding the clams, the conditions of the

storage described on this paper as well as the season of harvest were not significant. The survival of clams was high (5 deaths in 72 individuals) and so, the relationship with storage conditions was not significant ($p > 0.05$). The results of the sensory quality evaluation for oysters and clams showed a good quality for these bivalves subject to different temperatures and time of storage.

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List of Abbreviations

ADP - Adenosine 5'-diphosphate

AEC - Adenylic Energy Charge

AMP - Adenosine 5'-monophosphate

ATP - Adenosine 5'-triphosphate

QIM - Quality Index Method

TVB-N - Total Volatile Base Nitrogen

1. Introduction

Bivalves are invertebrate animals, protected by an exoskeleton in the shape of a shell with two valves closed through the adductor muscle. These animals are filter feeders, meaning that for breathing and feed, they filter large amounts of water in order to retain phytoplankton, microorganisms and organic particles that are suspended in the water (Silva et al., n.d.). Approximately three million metric tons of bivalves are harvested every year, being the main categories of oysters, mussels, scallops and clams (Morton, 2018). With the increasing human population in the world, the demand and the necessity for food is also increasing every year. Consumption fills this growing demand by humans as it is an important source of protein and fatty acids that bring many benefits to human health. Bivalves are a good alternative since are a great source of protein, omega 3, being also rich in minerals (Chen, 2011; Venugopal and Gopakumar, 2017). In addition, they are sustainable since are filter feedings, so "clean" the ocean, they are lower down in the food chain and, comparing to tuna or shark, contain less amount of heavy metals, which are toxic for humans (Cranford et al., 2013; Guy, 2016).

Although there is a growing demand for seafood, consumers are also increasingly selective towards the food products they eat (Jennings et al., 2016). The environmental impact caused by the growth or by the production of seafood is also an important factor in consumer decision, togther with safety and quality of the food product that will cause the consumer to reject (or not) the product or change supplier. People are becoming more informed and therefore more demanding with the quality of food that comes to their table and thus, farmers are increasingly concerned about how to maintain or improve their products to get the highest quality. However, quality is a very broad concept that depends on several factors, such as the preferences of the consumer in a particular place or, the age and the experiences of each individual. Moreover, the concept of quality is strongly linked to the conditions with which bivalves have grown, such as water salinity and temperature, food source, and season of reproduction (Kawashima and Yamanaka, 1992). For instance, it is during autumn when *Pecten maximus* reaches its maximum concentration of carbohydrates. Then, in the cold winter period,

due to the hibernation state and the development of the gonads, the scallops use their own source of energy, thus reducing the carbohydrate concentrations (Duncan, 1993). With the oysters, the period of hibernation starts when low temperatures during the winter predominate, decreasing their physiological processes and stopping feeding. In this way, no bacteria enter in their body and those that were there are digested and destroyed (Hunter et al., 1928). Vital processes are maintained primarily through the glycogen stores, obtained during previous seasons (Gage and Gorham, 1925). When water reaches 15 °C, the oyster *Ostrea edulis* start feeding again and also starts reproductive activity with increasing temperature (Matthiessen, 2008). These naturally occurring changes during the year lead to differences in the organoleptic characteristics of bivalves and the consequent variation in quality (Christophersen et al., 2008; Duncan, 1993; Idler et al., 1964).

Mostly people agrees that the freshness is essential for the quality of a food product (Green-Petersen et al., 2012) and so, it is generally approved that an alive bivalve is the definition of quality and therefore, it also represents better prices for the farmer (Overaa, 2001). While alive and out of their habitat, they are sensitive to the stress caused by both aerobic conditions and temperature changes leading to considerable quality losses. Handling, transport and storage are stressful conditions for the shellfish, due to reduced oxygen availability. The scallops - *Pecten maximus,* are not able to close the valves when they are out of the water, being more vulnerable to desiccation (Brand and Roberts, 1973; Cashmore et al., 1996; Duncan, 1993). In addition, the development of microorganisms and the accumulation of excretory waste products leads to a deterioration of physical conditions and ultimately leading to death (Cashmore et al., 1996; Duncan, 1993). The packaging used, the time and the temperature of both transport and storage are decisive factors in the variation of the organoleptic characteristics (Otoni et al., 2016). It is, therefore important to establish a limit time window for storage of bivalves as well as determine the best temperature at which they have to be stored.

Although it is an abstract concept, there are several ways to determine the quality of seafood. With the deterioration of the product, there are biochemical changes that can objectively defined the bivalves' quality. So, there, can be established: *1)* Levels of chemicals such as: lactic acid, and octopine for scallops, Adenosine 5'-triphosphate (ATP), Total Volatile Base Nitrogen (TVB-N), generated from the degradation of nitrogenous compounds by microbial activity or

post-mortem nucleotide catabolism (Boulter, 1996; Cao et al., 2009; Duncan, 1993; Jiménez-Ruiz et al., 2013; Kawashima and Yamanaka, 1992; Ruiz-Capillas et al., 2001; Zhang et al., 2017); *2)* The variation of pH is a deterioration indicator, and it is associated with the transformation of glycogen into lactic acid by fermentative bacteria (Montanhini and Neto, 2015); (Cao et al., 2009); (Zhang et al., 2017); *3)* The development of spoilage bacteria, since it is correlated with flavor changes, such as: *Vibrio* and *Aeromonas*, *Brochothrix thermosphacta*, *Pseudomonas* spp., *Shewanella* spp., and *Brochothrix thermosphacta,* are microorganisms which are involved in the bivalves deterioration (Coton et al., 2013; Duncan, 1993); *4)* A quality assessment can be made using the organoleptic characteristics of the product. Although it is a more subjective method, sensory evaluation is a simple an inexpensive method that can provide a good perspective of the consumer preferences. The evaluation of the quality by the sensorial assessment can be done through the evaluation of smell, appearance, flavour and texture of the scallops (Boulter, 1996; Coton et al., 2013; Makri, 2009; Ruiz-Capillas et al., 2001), oysters (Aaraas et al., 2006; Hasanspahić, 2011; Wang, 2015), and clams (Gonçalves et al., 2009; Torres, 2011).

So far, although there have been studies on the different biochemical characteristics during the different seasons (Dridi et al., 2007; Duncan, 1993; Matias, 2013; Ojea et al., 2004; Wang, 2015), there are no studies of the organoleptic differences which occur throughout the year. In addition, the quality of living bivalves may be influenced by temperature and storage time. However, no study, known by the author, compares the evolution of sensory parameters during different storage conditions with sizes of the oysters, clams and scallops in different seasons. Also, no previous study compared the survival during the storage with the quality of these bivalves.

In this study, I aim to check the effects of different storage conditions of alive scallops (*Pecten maximus*), oysters (*Ostrea edulis*), and clams (*Arctica islandica*) in the final product quality. To achieve this goal, I set up a verification method using, two different sizes of scallops, two different sizes of oysters and one size of clams, and evaluated which are the best temperatures and storage times. Also, I checked if mortality that can occur during the process influences the organoleptic characteristics that may or may not lead to rejection of the product. In

addition, in the final quality of these three bivalves was also compared the storage conditions with the seasonality, through sensorial evaluation.

1.1. Biology and life cycle of bivalves

1.1.1. *Ostrea edulis* (Linnaeus, 1758)

Biology

European flat oyster with the scientific name *Ostrea edulis,* belong to the phylum *Mollusca* and family Ostreidae (Perry and Jackson, 2017). These bivalves have two valves (one convex which fixes to the substratum and other which flat acts as a lid) and rough and scaly shells which have different shapes. Inside the shells are the meat which consists the mantle, gills, reproductive organs, adductor muscle and the circulatory, digestive and nervous system (He, 2000). The mantle is involved with the formation of the shell, and is also responsible for controlling the ingress of water. The gills are responsible for breathing function. Oysters feed through the mouth and allow the phytoplankton or other types of food particles to enter the stomach, where the mixture of enzymes perform the digestion (He, 2000). Adult oysters growth usually between 10 to 12 cm but can reach 20 cm (Gercken and Schmidt, 2014). Flat oysters live in intertidal areas often occur in beds on muddy-sand, muddy-gravels and rocks and can tolerate salinities of up to 23‰ (Svåsand, et al., 2007). During the winter, when temperatures are cooler, oysters go into hibernation, which means they stop feeding, slowing down physiological processes. Vital processes are maintained through glycogen stored during the active feeding season (Gage and Gorham, 1925). The microorganisms that are inside the body are destroyed by digestion (Hunter et al., 1928).

Ostrea edulis can be found from Norway to Morocco in the northeastern Atlantic, throughout the Mediterranean basin and also in eastern North America, from Maine to Rhode Island (Goulletquer, 2004) (Figure 1).

The species *Ostrea edulis* is threatened due to some threats such as overfishing, diseases, predators and due to some invasive species (Haelters and Kerckhof, 2009). The main predators are the invertebrates such as starfish, sea snails and crabs but birds and other fish can be included

(Encyclopedia Britannica, 2018). Also, some invasive species such as slipper limpet *Crepidula fornicate,* which degraded the oyster grounds or the American oyster drill *Urosalpinx cinerea*, which preys *Ostrea edulis*, are one of the causes of this species being threatened (Gercken and Schmidt, 2014; Haelters and Kerckhof, 2009).

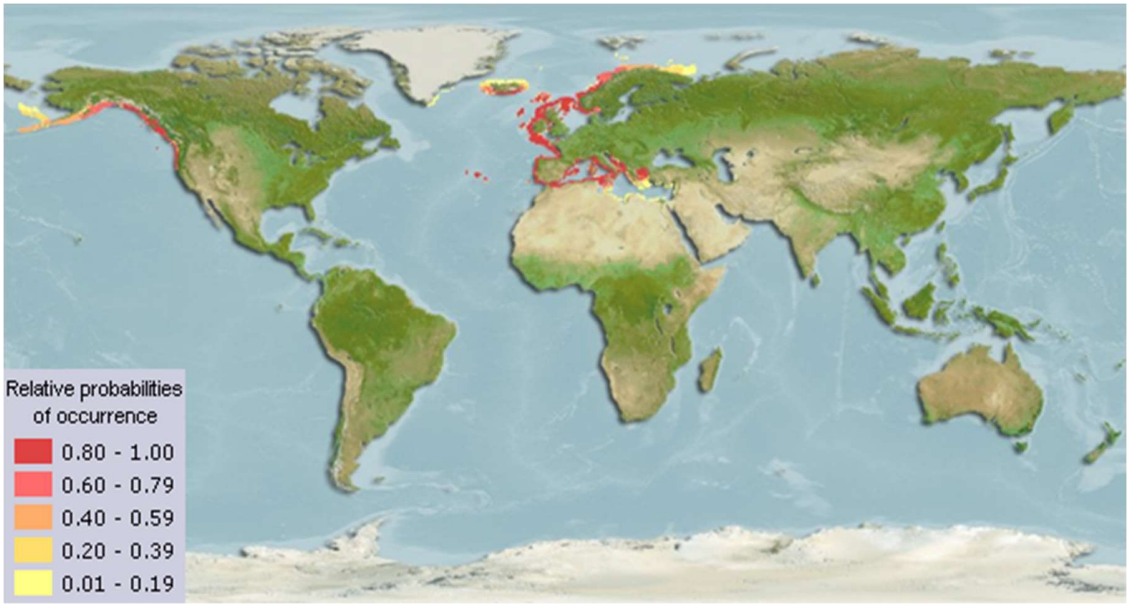

Figure 1: Distribution maps of *Ostrea edulis*. Distribution range colors indicate degree of suitability of habitat which can be interpreted as probabilities of occurrence (Source: Aquamaps, 2016a)

Reproduction

During the reproductive season, *O. edulis* changes sex twice which makes them hermaphrodites protandric (Goulletquer, 2004). Thus, they are males at the beginning of the spawning season, then they change to females and at the end of the season they change to males, influenced by the temperatures and the food supply (Gercken and Schmidt, 2014). However, these sex changes are related with the latitude and reproductive period since in Scandinavia, European flat oysters form one gender per year while in Mediterranean gender changes can occur several times in a year (Gercken and Schmidt, 2014). According to Svåsand, et al. (2007), the fertilization of European oysters occurs when about one million eggs are released and fertilized externally by the sperm. Fertilized eggs have 8-10 days of incubation period, and when the

formed larvae are released, they undergo to a stage of pelagic dispersion. This stage that can take 8-10 days ends when the larvae settle on the beds.

Harvest

Oyster harvesting can be done through hand-pick by divers, dredgers or pneumatic winches (Goulletquer, 2004). Harvesting by divers is a method more artisanal and environmentally friendly and causes less stress to the shellfish (Overaa, 2001). The dredges have steel teeth that crawl on the seabed, removing the oysters from their habitat to the bottom of the vessel. The suction dredgers function like a vacuum cleaner that pumps water from the seabed and therefore, also sucks the flora and fauna from the sea to the surface (Mercaldo-Allen and Goldberg, 2011). This is where the oysters are sucked, washed and sifted and then stored in the boats. These methods remove the bivalves as well as the fauna, flora and even the shells that are in the bottom. These changes have consequences for other oysters as there is a destruction of their beds. (Mercaldo-Allen and Goldberg, 2011).

The production of oysters had ups and downs reaching peak production in 1961 (29595 tons), decreased significantly during the following decade, and then recovered in 1970 (Figure 2). However, *O. edulis* was affected by two diseases throughout Europe, causing a drastic reduction in production. Despite all efforts, oyster production has never recovered to values once achieved (Goulletquer, 2004).

Figure 2: Global aquaculture production of *Ostrea edulis*. The graph shows the evolution of aquaculture production (tons) since the year 1950 until the year 2014. The year 1961 was the highest peak of aquaculture production (29595 tons) while 2011 was the lowest year of production (2173 tons) (Source: FAO, 2017a)

1.1.2. *Pecten maximus* (Linnaeus, 1758)

Biology

The king scallop, *Pecten maximus*, belongs to the genus *Pecten* and to the family Pectinidae (Morton, 2018). Like other bivalves, scallops feed by filtration through the gills by cilia. Through the water circulation, the microscopic plants and animals are captured in the gills, which also are the respiratory organ. They live on the bottoms, in clean sand, fine gravel, sandy gravel and sometimes mud (Svåsand, et al., 2007). The predators are starfish, crabs and in some cases, octopus, being the spats more susceptible to predation (Morton, 2018). The King scallop are found Eastern Atlantic Ocean, from North of Norway to Spain, and around north Africa, Azores, Madeira and Canaries (Figure 3).

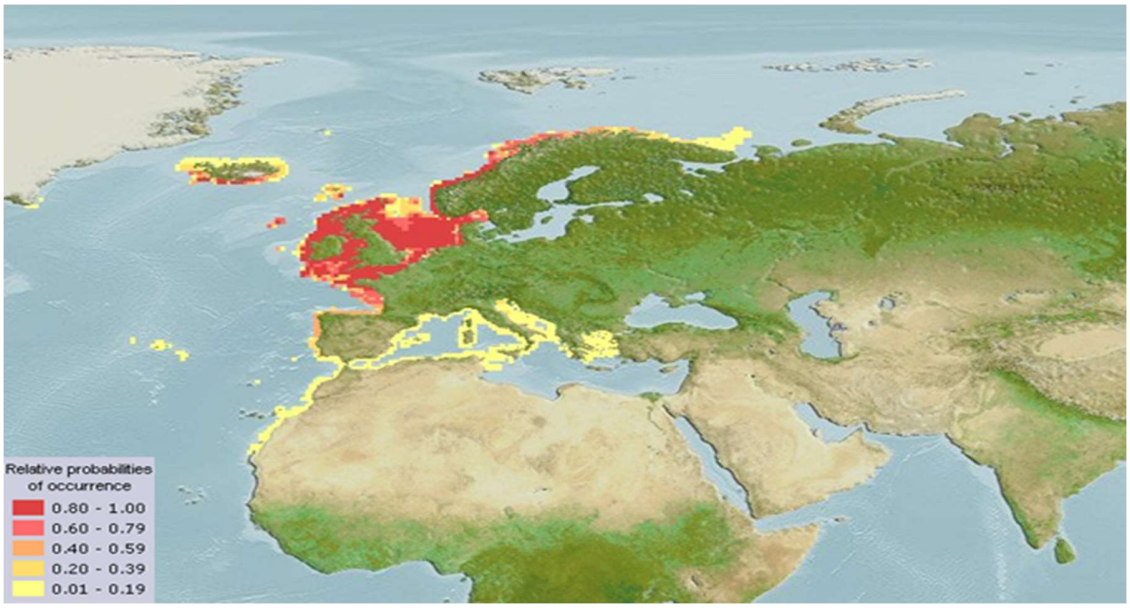

Figure 3: Distribution maps of *Pecten maximus*. Distribution range colors indicate degree of suitability of habitat which can be interpreted as probabilities of occurrence. (Source: Aquamaps, 2016b)

As the scallop grows, the mantle secretes the shell, forming a new ring every year. To reach the minimum commercial size (10-11 cm), it takes four years (Svåsand, et al., 2007). In the mantle (soft tissues which are in contact with the surface of the valve) are located the tentacles, which detect the chemical changes of the water, and several eyes, that detect the light (Morton, 2018). It has two valves united by a unique adductor muscle that allows the movement of opening and closing of the valves. If the scallops are disturbed, they react with a quick movement of the valves. Compared to other bivalves, scallops are very different in the ability to swim once they eject the water from the mantle cavity, pushing the animal forward due to spasmodic clapping movements of the valves (Duncan et al., 2016; Morton, 2018). Scallops react to the disturbance made either by predators or by harvesting by divers or fishing gear. This reaction is due to responses to variations in light, water vibrations and currents, which triggers movements of the neighboring scallops, due to the movement of an animal, causing mass movements. However, although the scallops are quick to react, the distances traveled are relatively short, and they quickly become fatigued, requiring some time to replenish energy levels (Duncan et al., 2016).

Reproduction

Pecten maximus is hermaphrodite, with a separate orange/red (in females) and white (in males) gonad. During spawning, eggs and sperm are released into the water. After fertilization occurs, the larvae can be carried by water currents to a considerable distance and then sink in the seabed. This dispersion, with different environmental conditions causes a great variability in the annual settlement of juveniles, with inevitable variability in the catch (Seafish, 2013). In the north, spawning occurs once a year but in south can occurs in several peaks (Svåsand, et al., 2007).

Harvest

Scallops are harvested in diverse ways in different countries. Traditionally, these bivalves are harvested using three different methods: trawling, dredging and diving. Trawling and dredging allow capture more quantities of scallops but has consequences on the environment. Dredges can have steel teeth which drag the seabed and then catch the scallops. Usually used in United States of America and United Kingdom, this method has a big disturbance on marine habitats, since it reduces the biodiversity and can bring invasive species (Seafish, 2013). Also, has a negative influence of scallops health and their habitat. Scallops which were harvest by dredging might presented damages on shells and have a lower ability to withstand the stress postharvest. Also, these methods have consequences in their habitat since removes and damage the organism that spat settle (Seafish, 2013). Due the unfavorable conditions of the seabed in Norway, dredging is not allowed in this country. The way that scallops are harvested is by hand-pick by scuba divers. Although this method is slow and expensive, allows to keep the quality characteristics of the scallops since it is not so stressful as the other methods (Overaa, 2001). And so, the Norwegian scallops are an exclusive product with high quality standards.

The scallop capture declined between 1970 and 1990. However, since the 90's, global capture are rising, reaching the maximum in 2012 (63 681 tons) (FAO, 2017b) (Figure 4).

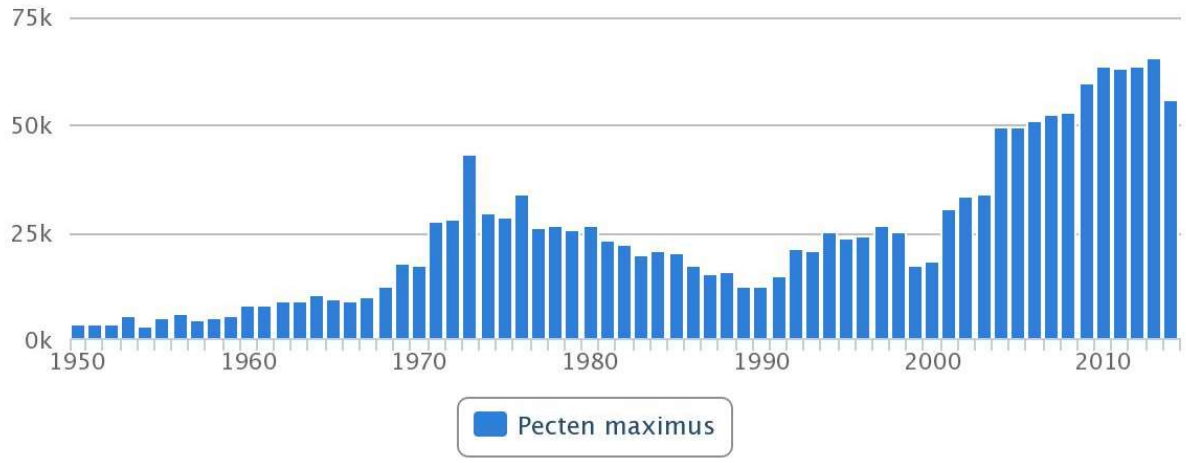

Figure 4: The global capture production of scallops, *Pecten maximus*. The graph shows the evolution of capture production (tons) since the year 1950 until the year 2014. The year 2012 was the highest peak of capture production (63 681 tons) while 1957 was the lowest year of production (4 700 tons) (Source: FAO, 2017b)

1.1.3. *Arctica islandica* (Linnaeus, 1767)

Biology

The ocean quahog, belonging to the family Arcticidae, are among the longest lifespan and slowest-growing marine organisms in the world (Sealife, n.d.). They have a short siphon and live preferentially buried beneath the surface of fine sediments and also between gravel sediments. These clams escape predators by burying themselves in the sediment while maintaining a stationary position (Stemmer, 2013). This behavior is due to the fact that this specie tolerates low concentrations of oxygen. The *A. islandica* can regulate its metabolic rate to the level of environmental oxygen as it accelerates the metabolism in oxygen rich environments or reduces the metabolism in the decrease of oxygen (Stemmer, 2013). The growing of the shell has influenced by the environment conditions such food availability, temperature and salinity. Also, the age and size of maturity may be dependent on growth rate and environmental conditions (Thorarinsdóttir and Steingrímsson, 2000).

The main threat to the species is large-scale trawling, but also changes in temperature and unintentional habitat, oxygen deficiency and mechanical damage (Sealife, n.d.).

The *A. islandica* can be found on Northern Atlantic and the Arctic, from Spain, north to Iceland, and from Cape Hatteras in North Carolina, USA to the Canadian Arctic (Sealife, n.d.) (Figure 5).

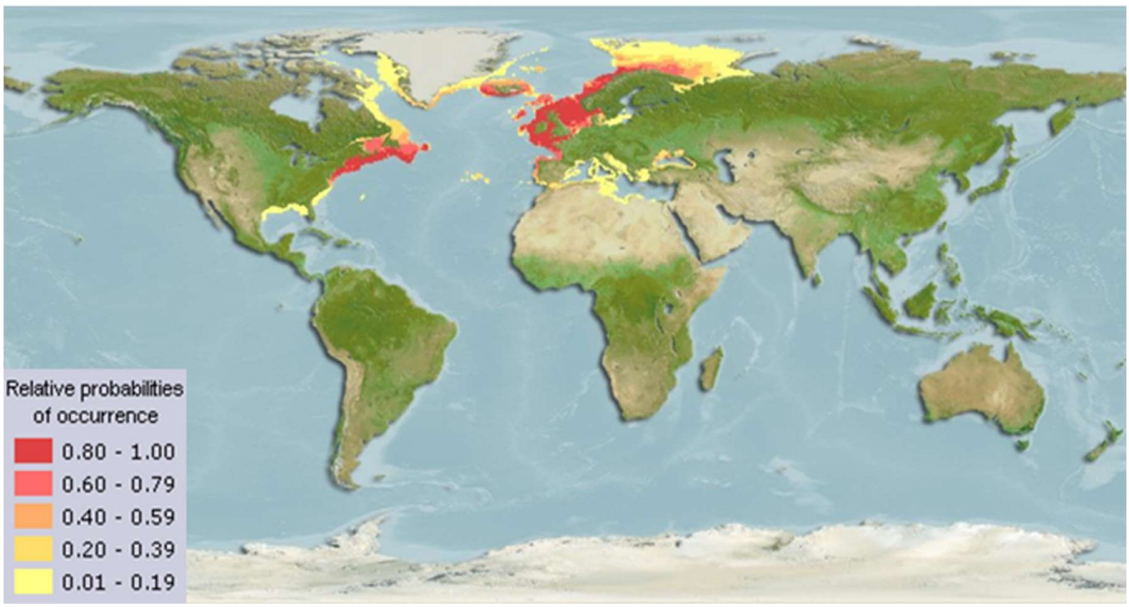

Figure 5: Distribution maps of *Arctica islandica*. Distribution range colors indicate degree of suitability of habitat which can be interpreted as probabilities of occurrence (Source: Aquamaps, 2016b)

Reproduction

Ocean quahog reaches maturity on a very late stage and can only begin sexual activity after 13 years in males and 12 years in females (FAO, 2018). Spawning, which is influenced by temperature, occurs from May to November (Mann, 1982).

Harvest

Ocean quahogs can be harvested through hydraulic dredgers that release high pressure water jets to loosen the clams from the sediments. The loose clams are picked from a mesh net bag that passes underneath the surface (Mercaldo-Allen and Goldberg, 2011). In relation to the global capture of *A. islandica*, the reported ranged never exceeded 200 000 tons. Since 1995 there has been a decrease in the total of the captures, with only a recovery in growth since 2002 (FAO, 2018) (Figure 6).

Figure 6: The global capture production of clams, *Arctica islandica*. The graph shows the evolution of capture production (tons) since the year 1950 until the year 2014. The year 1995 was the highest peak of capture production (185 881tons) while 1962 was the lowest year of production (440 tons) (Source: FAO, 2018).

1.2. Product Quality

It is difficult to determine a definition of quality, although it is a term widely used. It includes several aspects of a particular product, and varies from person to person. It is generally agreed that the concept of quality can be subjective and objective. It is subjective because it depends on the opinion of a particular person. Therefore, factors such as age, origin, lived experiences, etc., will dictate the person's preferences. For example, while in northern Europe, people are more opened to fish filleting and to the consumption of processed products (such as canned fish), in southern Europe consumers prefer fresh and whole fish such as sardines (Vanhonacker et al., 2013). The quality concept is also objective because it can be measured by food engineers and technologists through certain chemical, physical and microbiological parameters. Nielsen et al. (2002) argued that quality must be defined throughout the chain from the moment that the seafood is harvested to the consumer's plate and thus, it is necessary to establish quantitative methods of analysis that fit the entire food chain.

Karlsen (n.d.) described that quality can be defined according four main standpoints: variables in the individual product, production specifications, customer preferences and value (price of the product). The quality of the seafood may vary with the characteristics of the individual (such as species, gender and age), with external factors (such as where he/she lived, food, and water quality, temperature and salinity), as well as postharvest factors (such as the height of the year being harvested, harvesting method, stress during transport, handling and storage).

In relation to the aquaculture and fisheries worldwide, quality can be defined as the set of characteristics of each fish (muscle, skin, size, and age), ease of handling and/or processing as well as the environmental impact of the fish. However, it is accepted that the quality of seafood can be defined as "freshness" (Figure 7). Although fish have a certain level of "intrinsic" quality, this quality will always decrease after slaughter (Alasalvar and Taylor, 2002). Procedures that are carried out just prior to slaughter and post-slaughter will have a very negative impact on the quality. Thus, it is extremely important that pre-slaughter and post-slaughter processes are well considered and defined in order to keep as close as possible the "intrinsic" quality of the fish. In addition, there are some procedures that can be adopted post-harvest which can lead to the

improvement of seafood quality. For instance, besides the purification used to "clean" the bivalves of contaminants, this process can be used to improve the performance of these animals (Seafish, 1997).

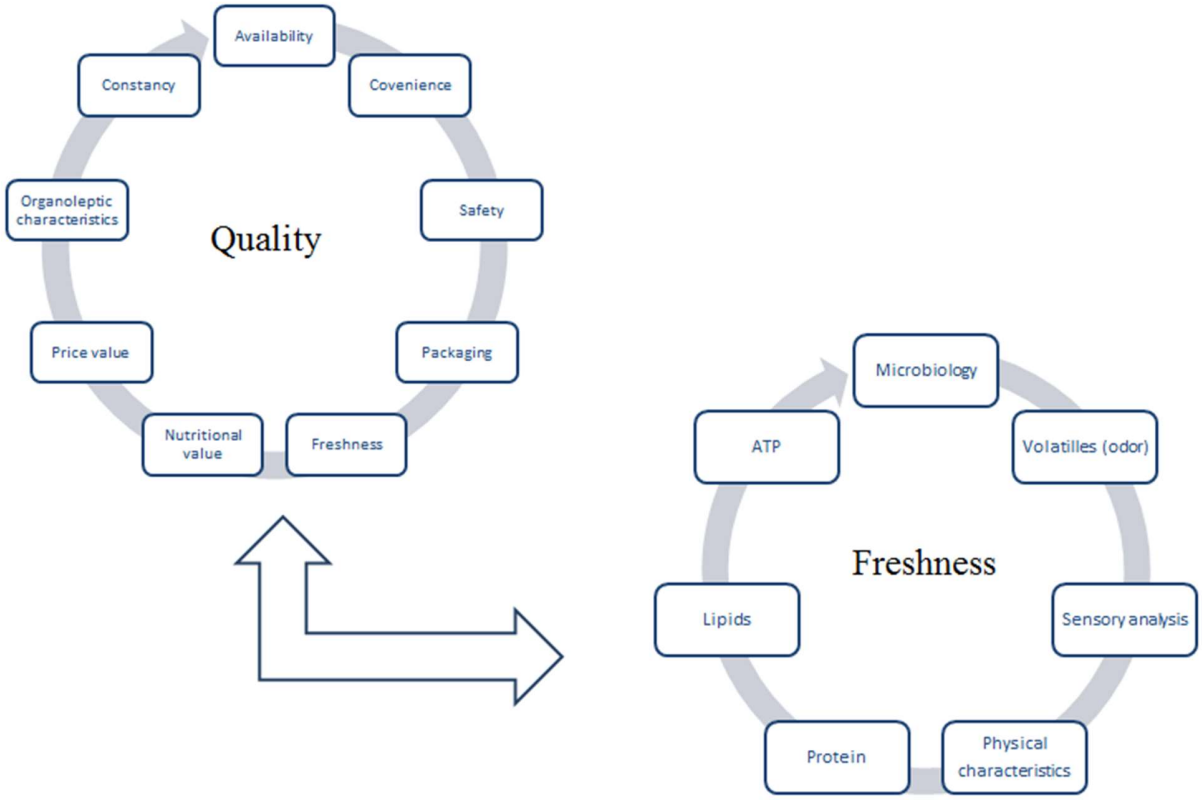

Figure 7: Relationship between the quality and freshness of the fish (adapted from Olafsdóttir et al. (1997). The "Quality" set encompasses factors that are related to quality. The set "Freshness" describes the parameters for evaluating the freshness of the fish.

1.2.1. Consumer preferences

Being the organoleptic characteristics among the most important ones when choosing a product by the consumers, their perception of quality is very important because it is their preference for a certain product that can dictate the sale and price of that product. Therefore, quality from the point of view of the consumer is of major interest to the food industry (from fishing/aquaculture industries to retailers). From a consumer perspective, quality can be defined by both sensory and physical characteristics of the fish, freshness, food safety, nutritional content, and ease of preparation. Regarding the sensorial characteristics, the aspect of the meat as the muscular texture or the color, can be a strong attribute that can lead to the rejection of the product. The case of salmonids is a good example. The flesh color of wild salmon is due to the consumption of crustaceans that the find in nature. However, the farmed salmon do not have access to this type of diet. To fill this gap, the industry adds a pigment, the astaxanthin, in the salmon feed to give the flesh of this fish its characteristic color. This pigment is an expensive ingredient, weighting around 15-25% of feed cost and 5-10% of total production cost (Waagbø, 2016). A study conducted by Steine et al. (2005) about the consumers' preference for red salmon showed that the redder the meat of the salmon was, the higher the price people were willing to pay for salmon. However, if consumers were informed of the origin of the red color, then their willingness to pay a higher price would decrease.

In France, the USA and Canada there is a practice of manipulating oysters in order to increase market value. While in France, the oysters that are in the final stage of growth are placed in specific places with the purpose of filtering phytoplankton-rich water in order to valorize the product, in the USA and Canada, overturn these bivalves or create them in rotating boxes to improve the characteristics of the shells (Cheney, 2010). It is also believed that the oyster acquires more flavor due to the fact that it repairs the shells more often and therefore increases the storage of glycogen.

In addition to the quality is directly related to the raw material, it is also strongly linked to the image and consumer confidence of the final product. Consumer behavior at the time of purchase is often influenced by brand and price, which are often important quality factors, especially when people are not able to make a good assessment of the quality of the product. Altintzoglou and Heide (2016) reported that the most involved and knowledgeable fish

consumers are those who are most concerned about the quality of the fish they buy and consume. On the other hand, the less involvement of people in the seafood area leads these consumers to value the price paid for fish more than the value of quality. Therefore, once the quality is achieved and its permanence is guaranteed, it is necessary to establish a simple and precise communication with the consumer in order to choose the product for its quality and not for other factors. Poor communication can make the consumer confused, brings it to mistrust the image of the brand. In a study conducted in the United States by Brayden et al. (2018), consumers showed preferences for wild products of unknown origin but when it came to shellfish and seaweed, they preferred certified products. Thus, the authors concluded that consumers had a tendency and even did not mind paying a higher price for the certification, and for the information on both the production and the origin of the products. Also, Manalo and Gempesaw (1997) reported that consumers did not mind to pay a higher price for oysters if they had, through inspection information provided by government entities, guarantees of food safety such as the assurance that oysters had been harvested in clean waters and that the post-harvest had been made following all the rules of quality and food safety.

While not all people are able to evaluate the freshness of the fish, the truth is that the unpleasant smell caused by nitrogenous and sulfuric compounds formed during the deterioration of seafood are well recognized by consumers. If in the past people once bought the fish directly from the fisherman, nowadays most of the consumers buy it at supermarkets and so, it is important to pass the information of the whole chain of seafood as well as to establish a strict traceability (Bremner, 2002).

Despite all the influences, the notion of food quality is ultimately influenced by consumer memories of previous experiences. It is during consumption that people obtain all the organoleptic characteristics of the seafood being integrated in their expectations regarding this product. In this way, different consumers will have different preferences since they will have different experiences with the consumption of seafood (Martinsdóttir et al., 2009).

1.2.2. Seasonal variations

In general, bivalves undergo biochemical variations during the annual cycle. During the winter, bivalves start a hibernation phase in which they use stored energies for biological maintenance. These energies, in the form of glycogen, are used both for the survival of the animal in the cold phase of the year and for gametogenesis. It is at this stage when the gonads develop and therefore require glycogen for the gametes. When the spawning season starts, glycogen content is generally minimal at the end of winter and maximum in autumn (Hummel et al., 1989).

Among the bivalve species, there is a high biochemical and seasonal variation depending on where the animals grow. According to Aníbal et al. (2011), there is a great seasonal biochemical variability of *Ruditapes decussatus*, due to the reproductive cycle. In their study, gametogenesis started in January, spawning occurred from June to September and resting state from October to December. Therefore, the authors concluded that the high nutritional values of the clams occurred in summer while the low values occurred in winter. A study conducted by Duinker et al. (2008), about the visual and tasteful evaluation of oysters *O. edulis*, showed that these bivalves get improvements in appearance, mineral taste and sweetness since September but it is in December that oysters reach fullness of taste, obtaining the highest scores in these parameters. With *Crassostrea gigas*, the glycogen levels and lipid levels were inversely related (Dridi et al., 2007). While in winter, lipid concentrations were minima and, glycogen levels were highest at this station. Lipids were accumulated in the gonads during the maturation period (spring) whereas glycogen, which has withstood the process of gametogenesis, reaches minimum values at maturity. Furthermore, the protein content was also higher during the maturation phase and decreased at the beginning of spawning (late summer). The authors concluded that this variation in fatty acid concentration and protein level were related to the availability of food (such as chlorophyll *a*). In this way, the condition indices values increases during this stage. Similar associations were found by Woll and Bakke (2017) who studied the seasonal variation of AEC levels (Adenylic Energy Charge) in the lion's paw scallop, *Nodipecten subnodusus*. AEC levels were lower during maturation of the gonads (September) than in the cold season. In sum, it is stated that the biochemical variability in bivalves, although it varies with species, is related to growth, with gametogenesis cycle, and to environmental conditions such as food availability and

temperature. These biochemical variations make the organoleptic qualities of these molluscs also vary throughout the year. Therefore, there will be seasonal preferences for specific molluscs by the consumer. As Aníbal et al. (2011) mentioned, the peaks of higher and lower nutritional value of the clams, *R. decussatus,* matched the peaks of greater commercial demand. Furthermore, the variation of the organoleptic characteristics occurs with several species of fish. In countries in southern Europe, sardines are much appreciated and are part of the gastronomy of these countries. In Portugal, the consumption of this fish is also linked to some traditional festivities at the beginning of the summer. It is this time of the year (June) when the sardine reaches the highest price due to the high demand. However, at this peak of demand, there are sometimes complaints from consumers claiming that sardines are "dry", flavor less, arguing that the body disintegrates when cooked. This phenomenon is due to the fact that the sardine has not yet accumulated the fat in the muscle, so much appreciated by the Portuguese people. The spawning takes place in the winter and so the sardine is accumulating fat, which is rich in polyunsaturated fatty acids (like omega 3), from spring to mid-autumn (IPMA, n.d.). This fat will be used in growth but also in the production of gametes.

In addition to the organoleptic changes which occur with bivalves at different times of the year, these animals have different responses to stress factors depending on the season. In the case of mussels, procedures prior to processing such as washing and declumping are harmful in the spring but was reported by Harding et al. (2004) that with *Mytilus* spp., these processes were beneficial in summer and autumn. In addition, it was concluded by Chandrapavan et al. (2012) that there were higher survival rates of scallops discarded during winter (+ 90%) than during summer (20-90%). Thus, thermal stress from large differences in seasonal temperatures was more critical to scallop survival than differences in scallop reproductive condition.

1.2.3. Stress

Stress can be defined as “an internal response of a living organism caused by environmental or other external factors that move that organism out of its normal physiological resting state, or homeostasis” (Selye, 1973). Stress causes an imbalance of the normal physiological state of the animal, forcing a reallocation of energy in the system. According to

Bartelme (2004), stress can be acute or chronic and mild or severe; therefore it is the severity, duration of stress and the health of the fish that will dictate the degree of this stress. Throughout the trade chain, such as harvesting, purification, transportation, and storage the bivalves are subjected to great stress factors such as oxygen deprivation and temperature fluctuations. Barrento et al. (2013) refereed that mussels are able to recover from these stressors, but if the factors are too intense, it will result in bad quality or even death. Bivalves react to stress in different ways. According to Widdows et al. (1979), *Mytilus edulis* close its valves and therefore the oxygen uptake is very low. Therefore, the anaerobic route is used, and the final products accumulate in the tissues. The same authors refereed that in the case of *Cardium edule*, the valves are opened in a stressful situation, maintaining a higher oxygen tension and therefore, a high aerobic rate. Such variability will condition both the handling and storage of bivalves. Therefore, it is very important to understand the behaviors of each species in a stress situation to establish criteria for quality assurance of the final product.

Depending on the degree and the duration of the stress period, the animal will become more fragile and therefore there will occur differences in metabolism, by decreasing the quality of the bivalve. Small stress inducers (such as salinity, temperature or drying) but induced during a long period may lead to the death of bivalves (Maguire et al., 1999). In addition, Schreck and Tort (2016) argue that if the animal survives, a prolonged exposure to stress may affect other vital functions such as growth, disease resistance or reproduction. Furthermore, these authors also mention that the way the fish respond to stress can differ greatly among species and within species, due to genetic differences. Thus, stress negatively affects the final quality of seafood, which causes negative consequences for the industry.

The characteristics that define the quality of the animal can lead to significant economic losses as for manufacturing companies or even lead to rejection of the product by the consumer. The stress that the fish suffers during harvesting, crowding, transport and handling influences the characteristics that are related to the quality parameters such as the texture and the color of the meat, and liquid leakage (Lacoste et al., 2001; Lerfall et al., 2015; Merkin et al., 2010; Refaey et al., 2017; Roth et al., 2009). The salmon industry is a good example of how stress before slaughter can influences the quality of the meat. The loss of texture of the flesh of *Salmo salar*, due to the massive accumulation of glycogen, causes soft fillets that are not suitable for the

manufacture of high-quality products (Torgersen et al., 2014). So, when this phenomenon happens, soft fillets are devalued leading to economic losses to the industries. Also, as already mentioned, the color of the meat is a strong attribute related to quality when it comes to the consumer choice. However, although industry wants to ensure quality, there are sometimes variations in the product. According to Alasalvar and Taylor (2002), fish with high levels of activity at the time of slaughter, like tuna, have lighter, less red and more translucent meat. This is also true with salmonids because stressful conditions during slaughter cause paleness in the muscle, reducing the positive attributes of fish quality. Glycogen is reported as an indicator of stress in fish as its content decreases with increasing stress. When, for instance, bivalves enter into stress, due for example to oxygen deprivation or temperature fluctuations, this compound is used during stress and as a result, there is the formation of organic acids, especially at higher temperatures. Thus, the glycogen is a good indicator of the physiological condition (Anacleto et al., 2013). Moreover, Hummel et al. (1989) reported that acetic acid is one of these acids and it is related with the mortality of *M. edulis*. The authors concluded that high concentrations of acetic acid appeared when there was a high mortality in the bivalves and therefore, it means that the acidification caused by the organic acids is catastrophic. Changing conditions after harvesting can bring benefits in the quality of the final product. Mørkøre et al. (2008) reported that if salmon, *Salmo salar*, is subjected to a five-week starvation period, the animal will better withstand acute stress before slaughter and therefore there will be improvements in meat firmness.

1.2.3.1. Effect of different methods of fishing

There are several methods of harvesting bivalves. These methods depend on either the type of bivalve or the type of seabed where these molluscs live on. In addition, the rules of each country may vary the methods of harvesting as well as the impacts the methods cause, both on the environment and on the final quality of the bivalves. The most environmentally friendly and least stress-inducing way is to catch bivalves by hand. The other methods of harvesting are by dredging and by trawling (Figure 8). These more intrusive methods have a greater environmental impact since the seabed and wild habitats may be destroyed. In trawling fisheries, fish caught in nets are trapped in the bottom with successive attempts to escape. This strenuous event leads

them to exhaustion and in turn to death. In addition, in the case of bivalves, these methods cause damages, sometimes in an irreversible way as they induce stress and damage the shells. It has been reported a mortality rate of around 15% of the scallops that are harvested by dredging an trawling (Caddy, 1973; Campbell et al., 2010). If death does not occur, the scallops during harvest experience a high stress which calls into question the quality of the final product. Maguire et al. (2002) studied the use of AEC and righting and recessing behavior. These authors concluded that larger scallops were less active and therefore with a lower AEC level than small scallops. Also, the dredging followed by emersion had an additional stress effect, although not sufficient to cause mortality in this study.

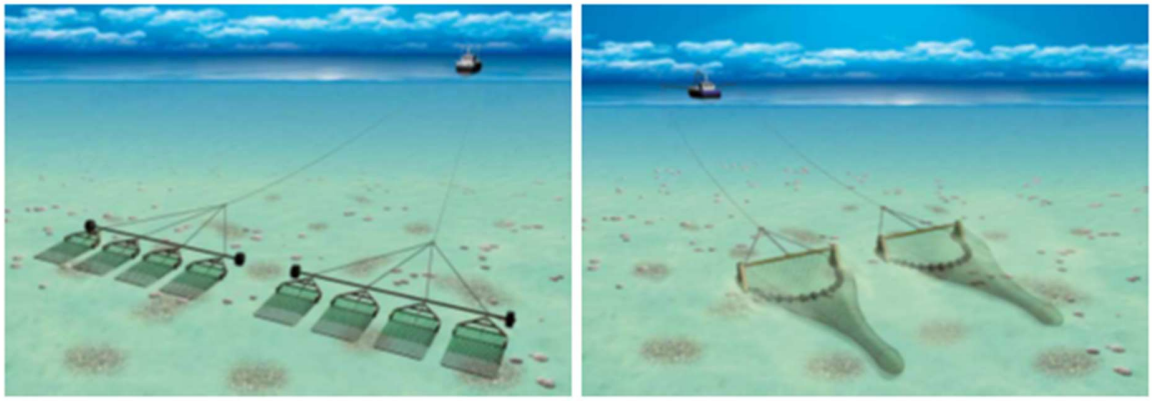

Figure 8: Different methods of harvest. On the right is the dredging method and on the right is the trawling method (Source: Montgomerie, 2015).

1.2.3.2. Handling, transport and storage

Wild bivalves have natural behaviors that make them unique in the animal world. While oysters are more stationary, scallops, through the opening and closing of the valves, can travel to other places to escape from predators, for example. They can also recess into the sediment. During storage in tanks, this behavior may not be possible due to the high densities of animals. In this way, the movements of the scallops can be conditioned due to the limitation of the opening and closing of the valves, causing stress in the animals. This phenomenon was studied by Maguire et al. (1999) and Woll and Bakke (2017), who concluded that there was a significant decrease in the quality of the scallops that were subjected to high densities. These bivalves

suffered chronic differences in carbohydrate content, recession speed and condition index and also, significantly affected the survival and subsequent transport.

There is a risk of mechanical shock throughout the entire processing chain of bivalves. During harvesting, the animals are placed in mesh bags or boxes in the boats, and they are then brought to the farm, where they can be placed directly in the refrigerator or tanks. The bivalves, which were inside the boxes or bags, are placed inside into the tanks and collected when necessary, handled and placed in packages for later transport. The transport phase involves loading, travelling (where there are always vibrations) and unloading. Throughout this process, there is a certain level of mechanical shock which can be detrimental to the final quality of the bivalves. Lacoste et al. (2001) concluded that mechanical stress made juvenile oysters more susceptible to pathogenic bacteria and favors an higher occurrence of mortality in these bivalves.

Bivalve transport and storage may be the most stressful steps of the entire chain for these molluscs. There are cases where transport is done in tanks and therefore the bivalves have access to oxygen through the aeration of the water. However, as this system is very expensive, it also involves many concerns about water aeration. Another type of transport is semi-dry or dry. The bivalves tolerate some time out of the water without losing quality. But the conditions in which they find themselves as the humidity, temperature and the time they are out of their natural habitat, will dictate the final quality of these molluscs. In addition to lack of oxygen, during dry transport or during dry storage, certain bivalves suffer of desiccation. This problem is considered one of the main environmental stresses for animals such as scallops, since it is the ability of these animals to control water losses that will determine life or death (Duncan, 1993). Desiccation is very harmful because upon drying the respiratory surfaces and therefore, decreases respiratory efficiency. Another problem is that bivalves accumulate toxic metabolic products. Bivalves produce ammonia as the main nitrogenous waste compound. When they are emersed, ammonia accumulates in the cavity of the mantle of the bivalves, causing damages in the cells and in turn lead to the loss of the physiological function (Duncan, 1993). In this way, it is very important to establish all the necessary parameters to avoid as much stress as possible of the bivalves during transportation and storage. The temperature has also a direct influence on the quality of the bivalves. Several studies as of the Cashmore et al. (1996) reported that high temperatures lead to increased energy demands, increase toxic products and accelerate bacterial development.

1.3. Quality assessment of bivalves during storage

Although it is a very broad topic, the quality of a food product can be measured through several parameters. During the storage of bivalves, there are biochemical changes that can serve as indicators for the industry to evaluate the quality of these molluscs.

Glycogen is processed by the bivalves through feeding during the fattening season and is then used as a source of energy during the hibernation phase for the maintenance of vital functions and development of the gonads. Thus, this energy reserve decreases during the gametogenesis and is lost by the adult during the spawning (Hummel et al., 1989). These changes in glycogen levels may occur during the natural or unnatural environmental changes to which bivalves are subject. Changes in temperature, salinity, starvation, anaerobiosis or even predators cause stress in these molluscs leading them to spend the energy resources (Jiménez-Ruiz et al., 2015). If the mollusc cannot cope with stress, due to its intensity or persistence, it can lead to the death of the bivalve. According to Hummel et al. (1989), prolonged exposure of bivalves to the air would bring animals to the same stress conditions as starvation and therefore leads them to use glycogen as the main energy source during exposure. This energy expenditure occurs suddenly coinciding with the death of the bivalve.

Adenylic energetic charge (AEC) levels in bivalves are also used for quality determination. AEC is calculated based on the relative abundance of adenosine tri-, di- and monophosphate (ATP, ADP and AMP), and gives a measure of available energy in the tissue analyzed (Woll and Bakke, 2017). This indicator was studied by Maguire et al. (1999) and Woll and Bakke (2017) with scallops that suffered a period of stress. The first authors indicated that in a healthy scallop the AEC levels would be 0.8 to 1, whereas that of a very stressed scallop the levels would fall to values between 0.3 and 0.5. Therefore, as a conclusion of both studies, it was mentioned that the more stressed the bivalve is, the more energy it uses and therefore the AEC level decreases.

One of the indexes most used to test the level of quality in seafood is the content of TVB-N. This parameter, resulting from the degradation of nitrogen compounds by the microbial activity can be used as the limit of bivalves' acceptability (Mota, 2013; Ruiz-Capillas et al., 2001). Zhang et al. (2017) reported very low TVB-N levels after the shelf-life of oysters. This

phenomenon may be due to the fact that the oyster converts glycogen to lactic acid, undergoing general acidification.

The weight loss and the values of the pH changes are two simple and inexpensive methods of measuring the evolution of bivalve quality. During the bacterial deterioration occurs the fermentation of the carbohydrates producing organic acids. This increase in acidification can be measured by the pH of the product (Cashmore et al., 1996; Mota, 2013; Zhang et al., 2017). Also Buzin et al. (2011), who studied the loss of water intervalval by oysters, concluded that when these bivalves leave the valves opened is due to the fact that the adductor muscle relax due to a certain stress that the oyster was subjected. This phenomenon leads to interval water losses and in turn to weight loss. The same conclusions were gotten by Ali and Nakamura (1999), who studied the air-breathing capacity of some bivalves, stated that there is a relationship between the differences in air-breathing rates of the species and the degree of shell opening. The authors demonstrated that the bivalves that had an upper aperture degree of the shells during aerial exposure showed oxygen consumption rates also higher than those with semi-closed or closed shells.

The determination of the microbial flora is a good tool to assess the evolution of the bivalves quality during the storage. The bivalves, being filter feeders, contain their own microbial charge. This microflora is more varied than in finfish, being related to the environmental conditions (Cao et al., 2009). The microorganisms' development causes the deterioration of the seafood that leads to losses of quality of the final product. Some microbial groups, such as Specific Spoilage Organisms, are responsible for the negative organoleptic characteristics such as unpleasant taste and smell in fish (Mota, 2013). The most common spoilage bacteria reported in fish and fish products are *Shewanella putrefaciens, Photobacterium phosphoreum*, lactic acid bacteria and *Pseudomonas spp*. (Goulas et al., 2005; Serio et al., 2014).

The organoleptic changes which take place during the storage of the bivalves can be determinant to verify the evolution of the quality of these molluscs. Sensory analysis, while being subjective, allows processors to assess the state of deterioration of the product while giving the perspective of the consumer to the industry. This analysis is widely used in seafood organoleptic evaluation and different methods can be used, depending on the research questions. The methods such as Quality Index Method (QIM), Torry Scheme and EU (EAB) Scheme are

widely used for the sensorial analysis of the fish. Torry Scheme is the most commonly used scale for the freshness evaluation of cooked fish (Olafsdóttir et al., 1997). The QIM method is focused for studying the impact of storage conditions on both the quality and shelf-life of raw fish (Hassoun and Karoui, 2015). EU (EAB) Scheme is used in ports to label the fish and is carried out by inspectors or sellers, not being sufficiently precise for the processing industry (Cooper, n.d.). The evolution of quality in bivalves can be carried out through the evaluation of parameters such as smell, appearance, flavour and texture of the scallops (Boulter, 1996; Coton et al., 2013; Makri, 2009; Ruiz-Capillas et al., 2001), oysters (Aaraas et al., 2006; Hasanspahić, 2011; Wang, 2015), and clams (Gonçalves et al., 2009; Torres, 2011).

2. Material & Methods

King scallops (*Pecten maximus*) *n* = 560, European flat oysters (*Ostrea edulis*) *n* = 469, and clams Ocean quahog (*Arctica islandica*) *n* = 135 were supplied by a Norwegian company "ScalMarin" (Svartevikvegen, Rong, Norway), with approved facilities for the storage of bivalves. These bivalves were collected from wild populations, and were harvested by hand by scuba divers in Hordaland, Norway. The bivalves were supplied in March (referred as a cold season), June (referred as the warm season) and August (late warm season). The evaluation of clams was not performed in August due to a lack of supply. All bivalves were in tanks with running clean seawater at ≈ 8 ºC (Figure 9) at the day of delivery.

Figure 9: Oysters and scallops stored in tanks with running seawater at the *ScalMarin* premises.

Although there were available five different sizes of scallops, for this experiment was selected only the smallest scallops and biggest scallops (with the width for small scallops: ≥ 10 cm ≤ 11 cm with the commercial name "Scallops superior", and for big scallops: ≥ 13 cm with the commercial name "Scallops XL"). Regarding oysters, there were five different sizes of oysters, and for this experiment there were only two sizes called "small oysters" (weight between 50gr – 70gr) and "big plus oysters" (weight between 110-150gr). This choice as due to the farmer of the *ScalMarin* related that smaller animals stay longer in the storage than bigger. To

make easier to identify these animals for this study, the scallops are called “small scallops” (for the “Scallops superior”) and “big scallops” (for the “Scallops XL”). The days of the loading, the bivalves were picked randomly from the tanks. Seven bivalves of same species with similar size were placed in a polypropylene box (one box for small size and one box for big size) with a wet sheet of newsprint placed over the animals to keep them wet, with any drainage. The oysters were placed tightly together to keep them under some pressure during storage (Figure 10).

Figure 10: Bivalves placed on a polypropylene box with wet newsprint in order to avoid dryness. Scallops (on the left) are only covered while the oysters (on the right) are completely involved with the newsprint.

Since the scallops are not able to close their valves out of the water as other bivalves do, their shells were closed with a clip to avoid dryness (Figure 11). The transport was carried on without any temperature control, and took no longer than 1 hour from the farm to the storage room. The storage, weight and survival assessment and, the sensory evaluation of all bivalves was done at University of Bergen premises (Norway), at BIO department.

Figure 11: Scallops to be stored already placed in a box with the clip in order to avoid dryness due to the opening of the shells.

The weather temperature was between 3 and 7 ºC in March, 16 - 24 ºC in June and 14 - 17 ºC in August. Once the boxes had been identified, they were stored in a different way: at different temperatures and during different days. Table 1 (for scallops), Table 2 (for oysters) and Table 3 (for clams) exemplify the way that the bivalves were stored in different rooms. All the boxes in all experiments were stored in the dark to mimic the storage conditions of an establishment that provides direct sales to consumers (Figure 12).

Figure 12: Bivalves being stored in a cold room. The light was switched on only when was needed to check or to pick up the boxes.

During handling of the scallops, it was observed in some samples, the presence of marine plants and small marine animals in boxes or even in the own shell of scallops (Figure 13).

Figure 13: Presence of strange organisms on the shells of scallops.

In the first experiment (March) there were 24 boxes of scallops (12 for small scallops and 12 for big scallops), 24 boxes of oysters (12 for small oysters and 12 for big oysters), and 12 boxes of clams. These 60 boxes divided by 3 different cooling rooms (3, 6, and 9 °C) in which 3 boxes of each species/size (3 of small oysters, 3 of small scallops, 3 boxes of the big oysters, 3 of big scallops, and 3 of clams) were stored during different days: 3, 6, 9 and 12 days (Table 1, Table 2, and Table 3). The weight of each bivalve was measured every 3 days in order to evaluate the changes in the weight that occur during the storage. Also, the survival of each animal after 3, 7, 10, 12 days of storage at temperatures referred above was checked. The survival was checked by immersing the animals in a tank with running seawater at ≈ 7 ºC, for 24h. It was possible to check almost all the living scallops in water since could it be observed they were capable of actively filter (tentacles out and moving). When there were doubts, the scallops were picked and the reactions and movements of the valves were checked when the mantle was touched. The animals which did not react and with opened valves were considered dead (Duncan, 1993; Maguire et al., 1999; Woll and Bakke, 2017). The survival of the oysters was checked after each oyster has been shucked. The gills and abductor's muscle were touched, and movements that proved their survival were checked (Aaraas et al., 2006). Regarding clams,

the survival was confirmed when movements of the edible part could be seen or felt after a mechanical stimulus (Mota, 2013).

As the smell of the scallops stored for 12 days was unpleasant and intense, it was concluded that it would certainly affect the participants, which would either lead to contamination of the air or to the confusion of scents by the participants. Therefore, in this way, there could be an erroneous evaluation of the remaining samples. Thus, in following experiments (March, June and August) were not performed the 12 days of storage of scallops. Moreover, due to the lack of supply of clams, was not performed both 3 and 6 days of storage in the winter experiment (March), the 3, 6 and 12 days of storage in the early summer experiment (June), and the whole late summer experiment (August).

Table 1: Demonstration of the arrangement of the scallops' boxes when they were stored in 3 different rooms, at temperatures 3, 6 and 9 ºC. In 1st session, 4 boxes (with small scallops) and 4 boxes (with big scallops) were stored during 3, 6, 10 and, 12 days at each temperature. In March (early summer), 6 boxes (3 of small scallops and 3 of big scallops) were stored at 7, 10 and, 12 days. In the June, 2 boxes of small scallops were store at 3 and 6 ºC, during 6 and 9 days and 1 box was stored at 9 ºC for 3 days. The big scallops were stored during 3, 6 and 9 days at 3 and 6 ºC and 1 box was stored during only 3 days at 9 ºC. Also, the boxes with the new cover and the boxes with absorbent were stored during 3, 6 and 10 days at 3, 6 and 9 ºC. In August, 4 boxes (2 boxes with big scallops and 2 boxes with small scallops) were stored during 3 and 6 days at each temperature.

Season	Session	Type	Room at 3 ºC				Room at 6 ºC				Room at 9 ºC			
March	1st	Small	3 days	6 days	10 days	12 days	3 days	6 days	10 days	12 days	3 days	6 days	10 days	12 days
		Big	3 days	6 days	10 days	12 days	3 days	6 days	10 days	12 days	3 days	6 days	10 days	12 days
	Winter	Small		7 days	10 days	12 days		7 days	10 days	12 days		7 days	10 days	12 days
		Big		7 days	10 days	12 days		7 days	10 days	12 days		7 days	10 days	12 days
June	Early summer	Small		6 days	9 days			6 days	9 days		3 days			
		Big	3 days	6 days	9 days		3 days	6 days	9 days		3 days			
		Cover	3 days	6 days	10 days		3 days	6 days	10 days		3 days	6 days	10 days	
		Absorbent	3 days	6 days	10 days		3 days	6 days	10 days		3 days	6 days	10 days	
August	Late summer	Small	3 days	6 days			3 days	6 days			3 days	6 days		
		Big	3 days	6 days			3 days	6 days			3 days	6 days		

Legend: - box with the 7 scallops

Table 2: Demonstration of the arrangement of the oysters' boxes when they were stored in 3 different rooms, at temperatures 3, 6 and 9 ºC. In 1st session, 4 boxes (with small oysters) and 4 boxes (with big oysters) were stored during 3, 6, 10 and, 12 days at each temperature. In March (early summer), 4 boxes of small oysters were stored at 3, 6, 9 and, 12 days and 3 boxes of big oysters were stored only during 6, 9 and, 12 days at each temperature. In June, 4 boxes (2 boxes with big oysters and 2 boxes with small oysters) were stored only during 6 and 9 days, at each temperature. In August, 6 boxes (3 boxes with big oysters and 3 boxes with small oysters) were stored during 3, 6 and 9 days at each temperature.

Season	Session	Size	Room at 3 ºC				Room at 6 ºC				Room at 9 ºC			
March	1st	Small	3 days	6 days	10 days	12 days	3 days	6 days	10 days	12 days	3 days	6 days	10 days	12 days
		Big	3 days	6 days	10 days	12 days	3 days	6 days	10 days	12 days	3 days	6 days	10 days	12 days
	Winter	Small	3 days	7 days	10 days	12 days	3 days	7 days	10 days	12 days	3 days	7 days	10 days	12 days
		Big		7 days	10 days	12 days		7 days	10 days	12 days		7 days	10 days	12 days
June	Early summer	Small		6 days	9 days			6 days	9 days		3 days	6 days	9 days	
		Big		6 days	9 days			6 days	9 days		3 days	6 days	9 days	
August	Late summer	Small	3 days	6 days	9 days		3 days	6 days	9 days		3 days	6 days	9 days	
		Big	3 days	6 days	9 days		3 days	6 days	9 days		3 days	6 days	9 days	

Legend:  - box with the 7 oysters

Table 3: Demonstration of the arrangement of the clams' boxes when they were stored in 3 different rooms, at temperatures 3, 6 and 9 ºC. In 1st session, were placed 6 clams at each box and 4 boxes were stored during 3, 6, 10 and, 12 days. In March (early summer) were placed 7 clams at each box and 2 boxes were stored for 10 days and 12 days. In June were placed 7 clams at each box and 1 box was stored for 9 days.

Season	Session	Room at 3 ºC				Room at 6 ºC				Room at 9 ºC			
March	1st	3 days	6 days	10 days	12 days	3 days	6 days	10 days	12 days	3 days	6 days	10 days	12 days
	Winter			10 days	12 days			10 days	12 days			10 days	12 days
June	Early summer			9 days				9 days				9 days	

Legend: - box with the clams

Winter – March experiment

On this session was carried out in order to evaluate their organoleptic characteristics – sensory assessment.

The scallops were store at 3, 6, and 9 ºC for 3, 7, and 10 days, and the sensory assessment was performed at the end of storage (Table 1). So, in total there were 9 boxes for small scallops (63 individuals) and 9 boxes for big scallops (93 individuals). During the session of sensory evaluation, it was observed that the scallops stored at 9 ºC after the day 6 presented very unpleasant odours. This event made the participants to take a bigger break in order to rest the olfactory sensation and for the efficient aeration of the laboratory.

Oysters were store at 3, 6, and 9 ºC during 3, 7, 10 and 12 days (small oysters) and during 7, 10 and 12 days (big oysters). The sensory assessment was performed at the end of storage

(Table 2). So, in total there were 12 boxes for small oysters (84 individuals) and 9 boxes for big oysters (63 individuals).

Clams were store at 3, 6, and 9 ºC during 10 and 12 days. The sensory assessment was performed at the end of storage (Table 3) and in total there were 6 boxes (42 individuals).

Early summer - June experiment

On this session was carried out in order to evaluate their organoleptic characteristics – sensory assessment.

Since the survival was higher with small scallops, was decided to store those animals only during 6 and 9 days, both at temperatures of 3 and 6 ºC. Regarding big scallops, they were stored at 3 and 6 ºC both for 3, 6 and 9 days (Table 1)). After the survival test, it was noticed that the scallops did not survive more than 3 days at 9 ºC and that even at that time almost half died (Table 2).

Therefore, it was also decided that at this temperature the scallops would be stored only for 3 days. In addition, it was hypothesized that perhaps the type of the scallops packaging influenced their quality after storage. Thus, additionally in this session, there were two more different ways of store the big scallops: 1) called “cover” - the scallops were covered with a paper intended to come into contact with food (previously moistened with seawater) instead of newsprint; 2) called “absorbent” - an absorbent pad, with superabsorbent polymers, supplied from the company “Absorbest”, was placed under the scallops to absorb all the liquids expelled by the animals during storage, keeping the wet cover of newsprint to avoid dryness (Figure 14). These two types of packaging were stored in three rooms at 3, 6, and 9 ºC, and at each temperature were stored for 3, 6, and 9 days (Table 1). In that way, were 5 boxes of small scallops (35 animals), 7 boxes of big scallops (49 individuals), 9 boxes of “absorbent” (63 animals), and 9 boxes of “cover” (63 individuals).

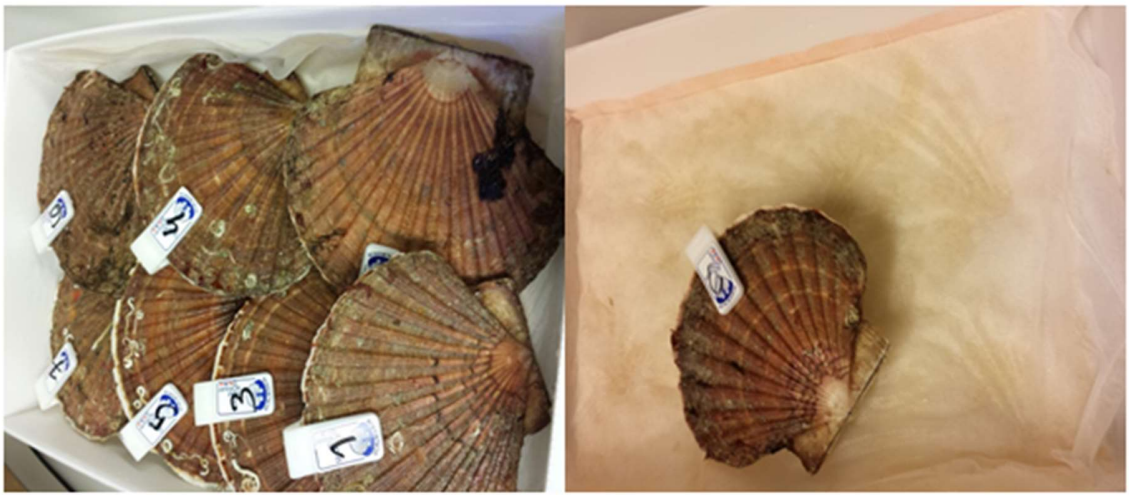

Figure 14: Scallops stored with an absorbent material in order to get the liquids released by the scallops.

The oysters were stored at 3, 6, and 9 ºC, during 6, 9 and 12 days (Table 2) and the clams were stored at 3, 6, and 9 ºC, only during 9 and 12 days (Table 3).

Late summer - August experiment

Regarding scallops, it was only performed the usual packaging with small scallops and big scallops stored at 3, 6 and 9 ºC for only 3 and 6 days (Table 1). Therefore, only 4 boxes of small scallops and 4 boxes of big scallops were stored (28 individuals each size).

Regarding oysters, 6 boxes (3 boxes with big oysters and 3 boxes with small oysters) were stored for 3, 6 and 9 days at 3, 6 and 9 ºC (Table 2).

For this study, the sensory assessment was performed by 10 participants: 4 of them on March, 3 in June and 6 in August (70% men, 30% woman). The tests were carried out at the lab of Biology department of the University of Bergen. Two animals of each species were randomly selected from each box, shucked and placed on half shell on an aluminium tray with a blind code. Moreover, each tray was covered with aluminium foil to avoid the dehydration. The trays were placed in a random order on a bench, totally covered, averting bias by order of presentation.

For the participants, water and crackers were provided to purge the residual smell notes between samples. Also, it was advised to take a break of 15 min every 4 trays, to minimize sensory fatigue.

Analysts were required to score the intensity of each characteristic describing the smell and the appearance. All the characteristics were evaluated by level of the intensity. The levels ranged since level 0 to level 5 (0 - absent, 1 – very weak, 2 – weak, 3 – medium, 4 – intense, 5 – very intense).

For the scallops, the sensory attributes were derived from some studies regarding the sensory profile of scallops (Archer, 2010; Boulter, 1996; Gonçalves, 2010; Maxwell-Miller et al., 1982; Ruiz-Capillas et al., 2001) and the following parameters were evaluated:

1) Smell – sulfhydric putrid, ammonia, sour, musty, boiled milk/seaweed, fresh /seawater;
2) Appearance – slight blackening, brownish color, slimy, bright surface.

For the oysters, the sensory attributes were derived from some studies regarding the sensory profile of oysters (Aaraas et al., 2006; Buzin et al., 2011; Cao et al., 2009; Hasanspahić, 2011; Lemasson et al., 2017; Otwell et al., 2011) and were evaluated the following parameters:

1) Smell – fresh/algal/seawater, sour, ammonia and, sulfhydric putrid;
2) Appearance – bright surface, body muscle dehydrated, plum white, plum well rounded, intact gills, intervalval fluid transparent and, intervalval fluid sufficient quantity.

For the clams, the sensory attributes were derived from some studies regarding the sensory profile of clams (Gonçalves et al., 2009; Torres, 2011) and were evaluated the following parameters:

1) Smell – fresh/algal/seawater, sour, ammonia and, sulfhydric putrid;
2) Appearance – bright surface, cream color, white color, brownish color.

In addition, in each evaluation, the participants were asked if the presented samples were suitable for eating or not.

Due to the limited budget for this experiment and due to not have professional panelists with enough knowledge about scallops in Hordaland, the sensory test was performed by experts in the scallop supply chain (fish farmer, scientist, and chefs), who work every day with bivalves and having good knowledge about the quality of these molluscs. Although some participants were the same on the three days of sensory assessment, it was not possible to repeat all the participants in all sessions due to their availability, since was the high season in their business.

3. Results

All the data were analyzed using the statistical software *Rstudio*. For analyzing both weight and survival were performed the linear models. Regarding the sensory assessment, due to having 4 predictors variables (2 continuous and 2 categorical variables) and the response variable being categorical was performed a linear regression model. A *posteriori* Tukey HDS test was used to contrast treatments. For the data of the question (if they would eat or not the bivalve) it was performed the generalized linear model. The level of significance was set at 95%.

In order to be easier to identify and analyze data, all storage days were identified as 0, 3, 6, 9, and 12 days. Thus, in some experiments, when the bivalves were stored for 7 and 10 days, the data analysis will be indicated as 6 and 9 days, respectively.

3.1. Oysters

3.1.1 Weight assessment

Although oysters lost weight during storage time, the weight losses differ on these bivalves (Figure 15). Small oysters lost less weight than big oysters. The small oysters lost 4% at 3 °C, 6% at 6 °C and 7% at 9 °C of their weight during storage. The greatest weight loss was at 9 °C for 10 days (7%). With the big oysters was different. The higher loss weight was with oysters stored for 12 days but at 6 ºC (20%). The storage at 6 ºC was the temperature which occurred more losses of weight, with the exception of the storage at 6 days, which was the 9 ºC (9%). Thus, in storage at 3 ºC the big oysters lost 10% of their weight, at 6 ºC they lost 20% and at 9 ºC there were 12% of losses of the total weight of all the big oysters. The day of the storage was statically significant ($p < 0.05$). The size and temperature were not significant ($p > 0.05$).

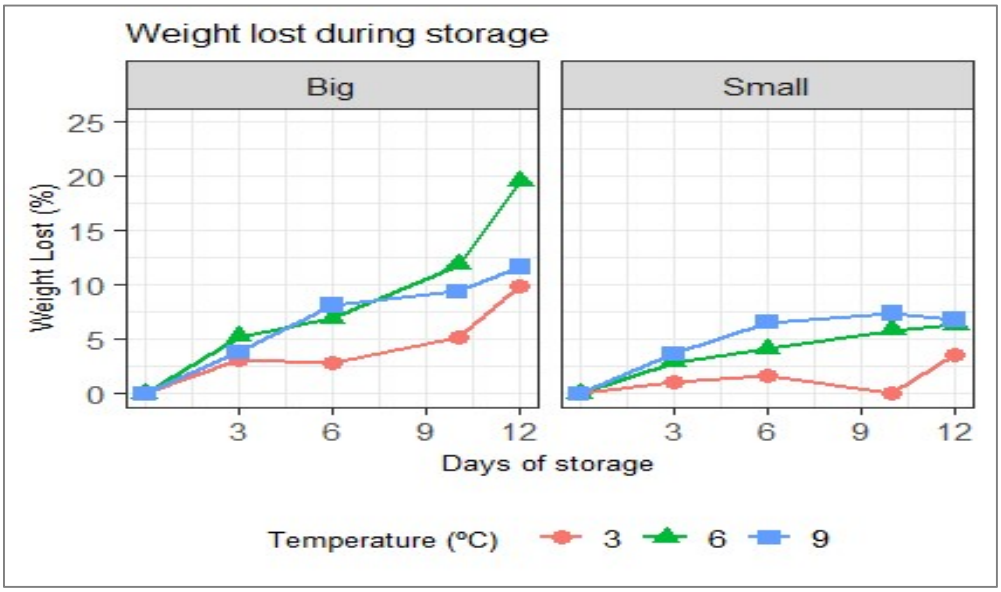

Figure 15: The weight lost (%) of oysters during the storage at different temperatures (3, 6, and 9 ºC).

3.1.2 Survival Rate

There were 6 deaths in 168 oysters during the storage (Table 4). At 3 ºC, 1 big oyster and 1 small oyster died during the first three days of storage, and 1 small oyster died in 7 days. At 6 ºC, 2 small oysters died (3 days and 12 days respectively). At 9 ºC, only one small oyster found death in 7 days of storage. The results showed that the survival of oysters stored during 12 days was not statistically significant ($p > 0.05$).

Table 4: Number of oysters which survived during the storage at different temperatures (3, 6 and 9 ºC). Each sample had a total of 7 oysters. During the storage until 12 days, there were 5 deaths of small oysters while there was 1 death of big oysters.

	Oysters small			Oysters big			
Temperature / Days	3 ºC	6 ºC	9 ºC	3 ºC	6 ºC	9 ºC	Total (survivors/total oysters)
3 days	6	6	7	6	7	7	39/42
7 days	6	7	6	7	7	7	40/42
10 days	7	7	7	7	7	7	42/42
12 days	7	6	7	7	7	7	41/42
Total (survivors/total oysters)	26/28	26/28	27/28	27/28	28	28	168

3.1.3 Sensory assessment

In total, 11 attributes were evaluated by the panel during the sensory test sessions.

Fresh, algal seawater: parameter associated with fresh oysters, was only noticed by the assessors as bellow medium of intensity (intensity level = 3). The p was > 0.05, therefore, this parameter was not statistically significant. In June, it was only reached the weak level of intensity (intensity level = 2) by all big and small oysters at 6 and 9 ºC, except the small oysters stored at 9ºC in 6 days of storage (which reached the medium level). There were oysters which this parameter was absent: in March, the small oysters stored at 9 ºC during 12 days of storage (in March), the big oysters stored at 6 and 9 ºC during 6 days of storage and, in June with the big oysters at 6 ºC during 9 days of storage (Figure 16).

Sour: It is the first parameter which is considered negative in the product low quality and therefore, should not be present. In this way, levels of intensity higher than "very weak" (intensity level = 2) mean poor quality. The p< 0.05 means that this parameter was statistically significant. In all seasons, the intensity of this parameter was evaluated bellow 2 (weak) for oysters stored at 3 ºC in both sizes, except small oysters stored in June in 9 days of storage,

which reached level 3. At 6 ºC, although the small oysters were always below 1 (very weak), with big oysters was very different since, in 9 days of storage, big oysters stored in March and June reached the intensity level 4 (intense) and the intensity level 3 (medium) respectively. August was the season which had lowest levels of intensity of Sour (between level 0 to level 1) (Figure 16).

Ammonia: this is another undesirable parameter in an oyster and therefore should not be felt by the consumer. In this way, values above the intensity level = 2 (weak) can lead to the rejection of the product. This parameter, with the $p > 0.05$ shows that Ammonia was not significant. Almost all the oysters were evaluated with the low levels of Ammonia (between level 0 to level 1), with the five exceptions: oysters stored in March at 9 ºC in 12 days of storage for small oysters (intensity level ≈ 3) and 9 days of storage for big oysters with level ≈ 4; small oysters stored at 3 ºC in 9 days of storage; big oysters stored at 6 ºC in March in 6 days, and June in 9 days of storage. Overall, August was the month with the lowest intensity levels. Strangely, in March greater ammonia smell was noticed with big oysters in 6 days of storage than in oysters stored in both 9 and 12 days, at 6 and 9 ºC (Figure 16).

Sulfhydric, putrid: It is the kind of smell which does not want to feel when someone is going to consume seafood. This parameter might be at similar levels as sour and ammonia since all are noticed due to the deterioration of the product. Sulfhydric, putrid was statistically significant since p was < 0.05. As the parameter of Ammonia, the intensity of Sulfhydric, putrid was very weak or absent with all the oysters in this study (Figure 16). However, there were four exceptions: with the small oysters, the intensity was almost intense (intensity level ≈ 4) at 9 ºC, in 12 days of storage in March, and medium (intensity level = 3) at 3 ºC in 9 days of storage in June. With big oysters, this parameter was almost intense (intensity level ≈ 4) for those stored for 6 days both at 6 and 9 ºC. Oddly, the values dropped to very weak (intensity level = 1) or absent (intensity level = 0) on the remaining days of storage. Thus, the month that presented the worst oysters in terms of quality was March (for small oysters stored at 9 ºC and for big oysters at 6 ºC and 9 ºC) and June (for small oysters stored at 3 ºC). Overall, August was again, the month with the lowest intensity levels (between intensity level 0 and intensity level 1).

Bright surface: this attribute is one of the most desired in oysters because it defines a fresh oyster, just shucked. There were few differences between the two oyster sizes, with the

results remaining under all storage conditions around the medium intensity (Figure 16). However, there was one exception in the small oysters in June because after 3 and at 9 °C for 9 days of storage, the brightness of surface was described as very weak (intensity level ≤ 1). Big oysters had the highest peak Bright surface in June at 9 °C for 6 days storage (intensity level = 4). This parameter was statistically significant *(*$p < 0.05$) in quality of oysters.

Body muscle dehydrated: this parameter is the opposite of the above, defining an oyster that is losing qualities. This parameter rarely exceeded level 2 (intensity level = weak), expected in small oysters at 9 days of storage at 3 and 9 °C (June) and at 3 days at 9 °C (August), and in big oysters for 7 days storage at 6 °C (June). Since the $p > 0.05$, this parameter was not statistically significant (Figure 16).

Plum white: with $p < 0.05$, this parameter was statistically significant. The intensity of this parameter in both March and in June was around the medium level (intensity level ≈ 3) for small oysters and above 3 for big oysters. However, it can be noticed that in August, the levels of this parameter decreased in both sizes of oysters: for small oysters, the levels were almost all between 1 and 2, while for big oysters, the levels were between 2 and 3 (Figure 16).

Plum well rounded: this parameter was statistically significant ($p < 0.05$). In August was the month which presented the lowest intensity (intensity level ≈ 2 for small oysters and intensity level ≈ 3 for big oysters). In March and June, the intensity of this parameter was between 3 and 4 for big oysters. For small oysters, the highest level was reached at the temperature 6 ºC in March in 12 days of storage (intensity level = 4). In June, the oysters stored at 9 ºC got were those which got more intensity, especially those of 9 days if storage (Figure 16).

Intact gills: with the $p < 0.05$, this parameter was statistically significant. In the case of big oysters, the intensity of this parameter was considered medium to intense (Figure 16). However, this parameter was evaluated as weak (intensity level = 2) either in August, in both 3 days and 6 days of storage at 9 °C, and in June for 6 days of storage at 6 °C. However, at this temperature, an increased stress with storage days. For small oysters, the level was gotten at 6 ºC in March in all days of storage (intense to very intense). Once again, there was a decrease in intensity during the days of storage. A peak reached the intensity level = 4 (intense) in March

and June after 9 and 6 days of storage, respectively, followed by a drop in intensity in March after 12 days (at 3 and 9 ° C) and in June at 9 days at three storage temperatures.

Intervalval fluid transparent: This parameter is connected to a fresh oyster and therefore its intensity should be high. The $p < 0.05$, this parameter was statistical significance. In the case of small oysters, the intensity was almost highest at 6 ºC during all the seasons (except at 3 ºC after 12 days of storage in March and after 9 days in August). March was the month with the highest levels (intensity level ≈ 3). With big oysters, the highest month was in August (intensity level > 3). In August, at 9 ºC, the intensity was around 3 (medium) after 3 days of storage, lowered to the level of weak (intensity level = 2) after 6 days, and then oddly returned to medium for 9 days of storage. This type of event also occurred in June, but at 9 ºC, with the intensity level around 2.5 (weak) after 3 days of storage, increasing to around 3.5 (medium) after 6 days, dropping to around intensity level = 1.5 (very weak) after 9 days of storage (Figure 16).

Intervalval fluid sufficient quantity: in June after 6 days of storage, the oysters which were stored at 6 and 9 ºC decreased the intensity at 6 and 9 ºC. However, until 6 days of storage, this parameter raised at 9 ºC. With small oysters, the temperature which got the highest levels was 6 ºC, being in March when they reached the highest intensity (intensity level > 4 = intense). In relation to the big oysters, March and August the intensity levels of this parameter were around the medium level, as in June only at 9 ºC. In this month, at 6 ºC the level of intensity was medium in 6 days of storage and decreased to weak in 9 days (Figure 16). This parameter was statistical significantly since $p < 0.05$.

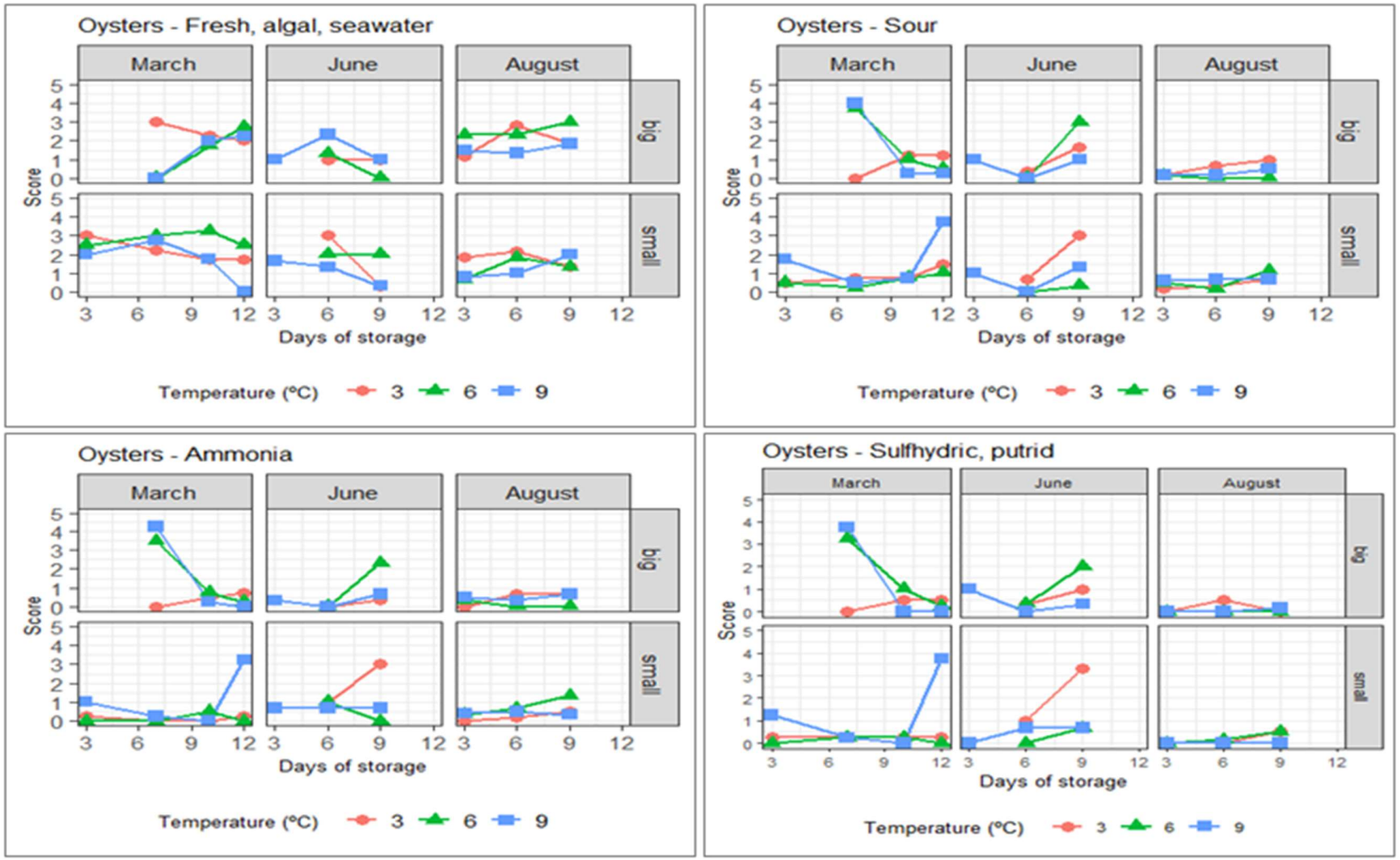

Figure 16: Average of the sensory evaluation of oysters during the storage, at different temperatures (3, 6, and 9 ºC). All the characteristics of the samples were evaluated by the participants with a level of the intensity ranged from the level 0 to the level 5 (0 - absent, 1 – very weak, 2 – weak, 3 – medium, 4 – intense, 5 – very intense).

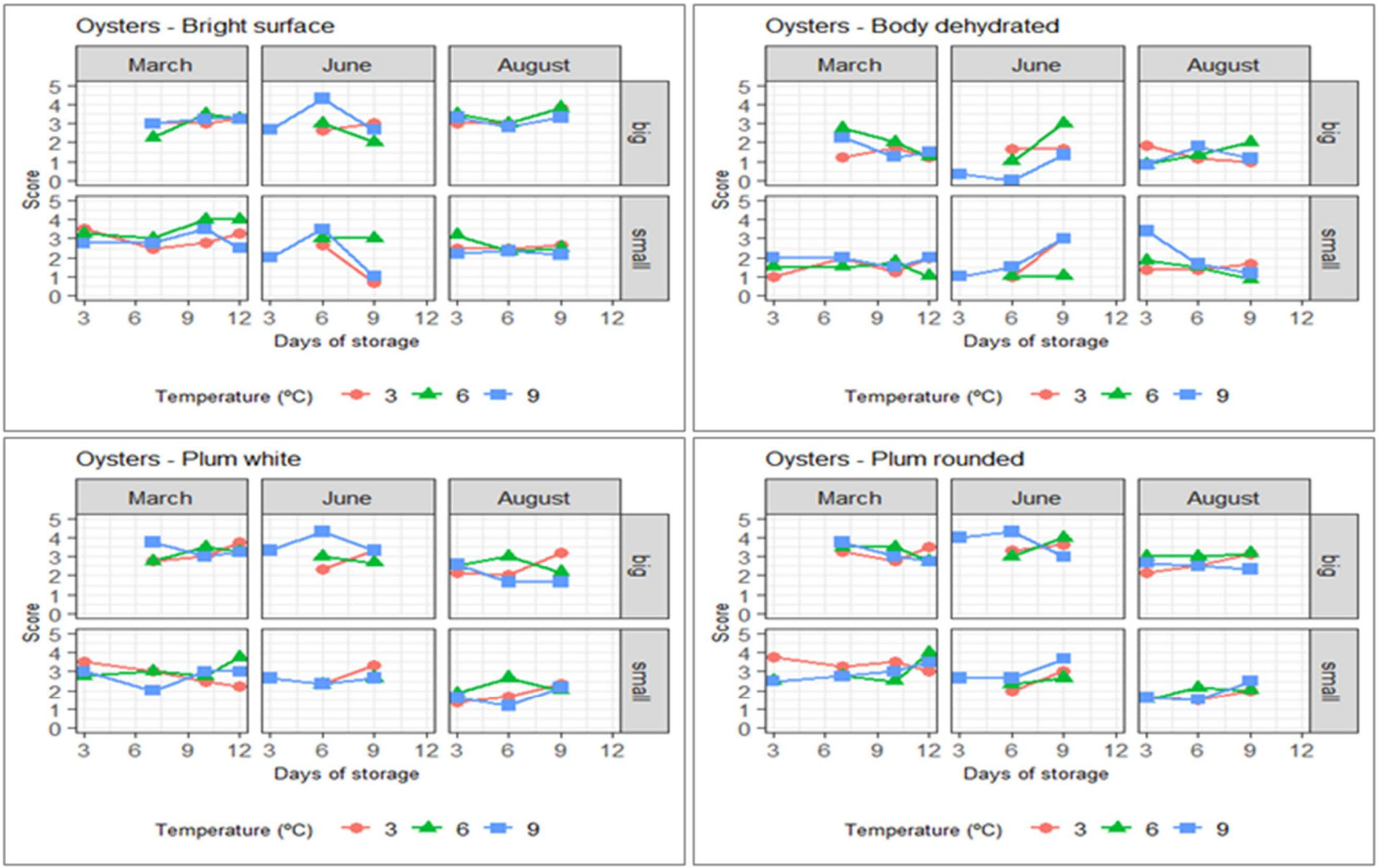

Figure 16: cont.

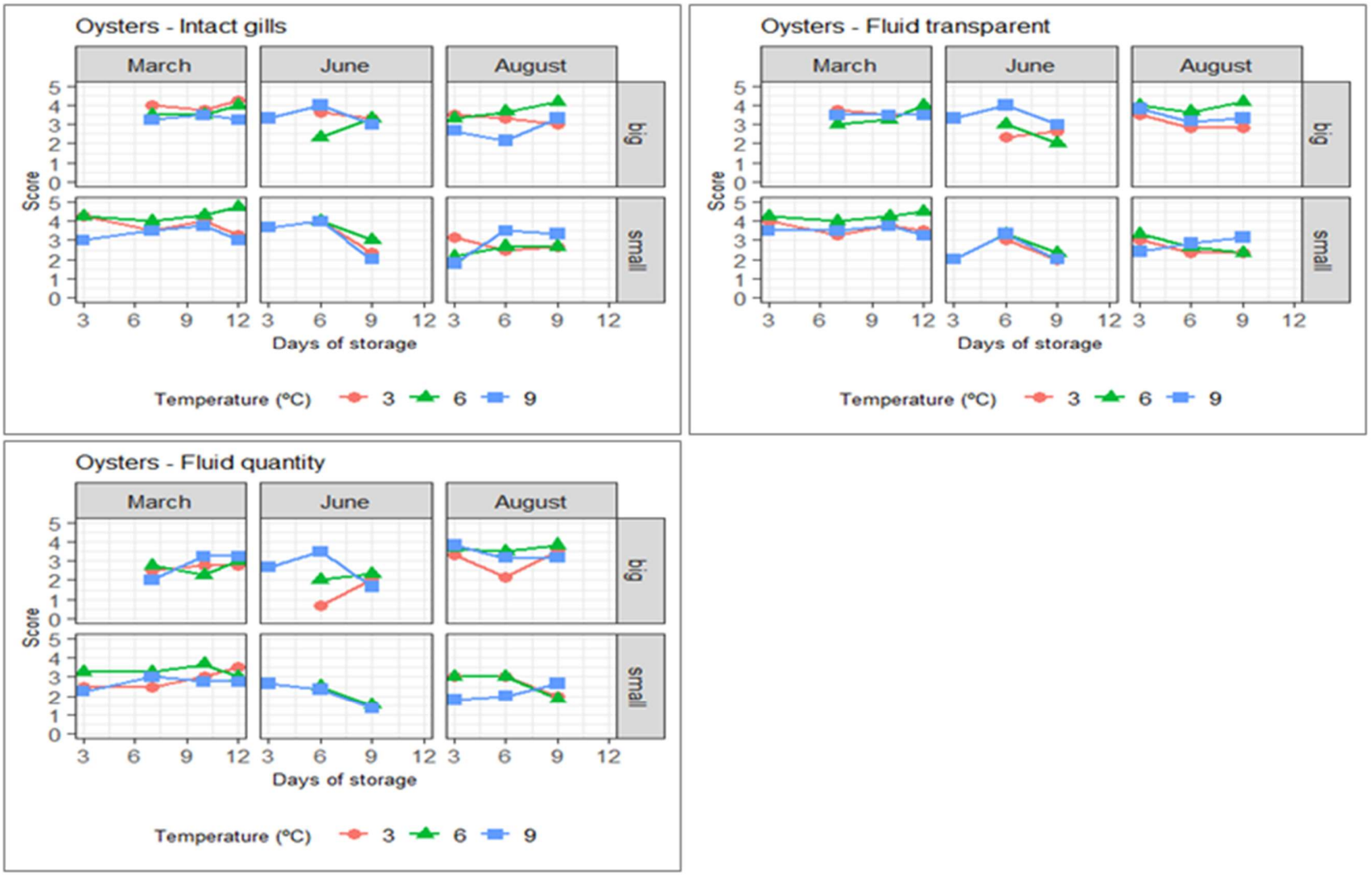

Figure 16: cont.

3.1.4 Acceptance of oysters

During the sensory evaluation, the participants were asked if the oysters' samples, which they were evaluating, appeared to be in good quality to be eaten (Figure 17). Small oysters were accepted by 100% of the participants only in some days in March: 6 and 9 days for all temperatures and 12 days for 6 ºC. In the same month, there was a difference in oysters at 12 days at 9 ºC because no one accepted the oysters under these storage conditions. The other small oysters rejected were those stored for 3 days in June, at both 3 and at 6 °C, and both 9 days at 3 and 9 °C. In June, almost all small oysters were discarded except for those that were stored in 6 days at all temperatures, which had an acceptance of 66.7%.

Regarding the big oysters, at 9 ºC almost all the oysters were accepted by the assessors with the exception of oysters to 3 days in June and 6 days of storage in March. The oysters stored at 6 ºC were rejected by all participants at 6 days on March, 3 and 9 days in June. However, they were accepted by 100% of participants for 12 days in March, 6 days in June and 83% on all storage days in August. For big oysters stored at 3 ºC, only those of 6 days in March were accepted by all participants. In the same month, only 25% of participants rejected 9 days but 12 days were rejected by 75%. In June, the oysters were all rejected in 3 days. August was the most positive month in the acceptance of the big oysters, being accepted by more than 70% of the participants.

The differences in the acceptance of the oysters between the seasons were significant ($p < 0.05$). Moreover, the differences between the small oysters harvested both in June and in August were significant ($p < 0.05$).

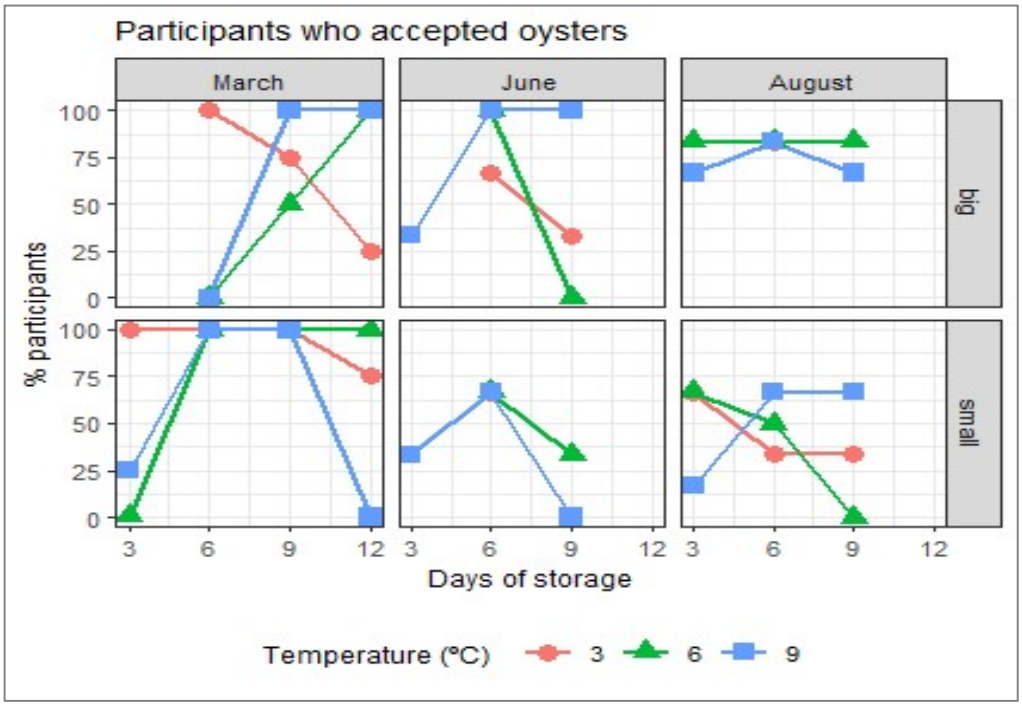

Figure 17: Acceptance of oysters by the participants**.** The number of participants who accepted the small oysters and big oysters is in percentage (%). In March, were carried out samples up to 12 days with small oysters and during 7, 10 and 12 days of storage with the big oysters, at the three temperatures. In June, the oysters were stored only 6 and 9 days at the three temperatures. In August were performed samples during 3, 6 and 9 days of storage at the three temperatures.

3.1.5 Quality level

When all parameters mentioned above are met, a method for evaluating the quality of the oysters during storage at a given time at a given temperature can be obtained, with an overview of quality at three seasons. The graph below is the result of an average proportion of the intensity levels of the positive and negative parameters in order to achieve a quality determination of the oysters studied in this paper (Figure 18). The level "0" is the lowest level of quality. The levels below "2", means that the oysters have poor quality and should be rejected. The levels between "2" and "3" are the medium level while the levels above "4" represent the best quality. Looking at the graph, it seems that August was the month that registered the least variability, whereas the month of March was the one that obtained the most. However, using Tukey HSD to compare the groups, seasons were not statistically different from each other ($p > 0.05$). Almost all the quality levels are between level 3 and level 4, except the oysters stored in 6 days at 3 and 9 ºC. However,

there was no significant variability of oyster quality throughout the storage time at three different temperatures, at three different seasons ($p > 0.05$).

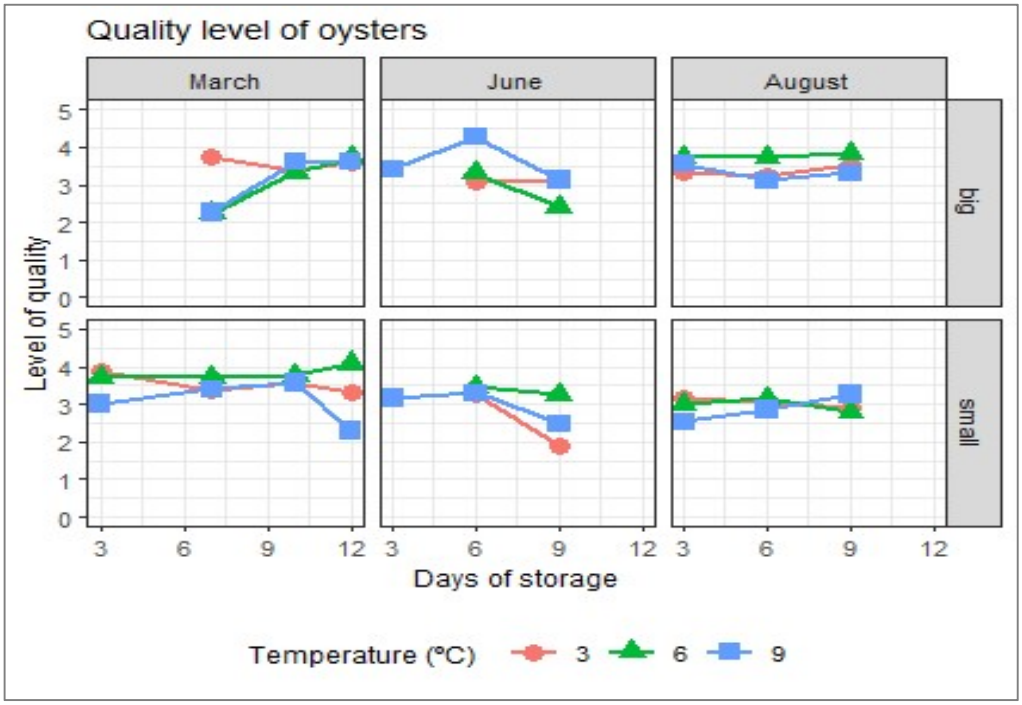

Figure 18: A quality overview of all oysters studied in this paper. The graph shows a ratio between negative and positive parameters, representing an overview of the quality of all oysters during storage at 3, 6 and, 9 °C, up to 12 days in March and up to 9 days in June and August. The level "0" is the lower level of quality. The level between "2" and "3" is the medium level while the levels above "4" are the best quality.

3.2. Scallops

3.2.1 Weight assessment

In this study, it was verified that the weight of scallops was influenced by storage conditions ($p < 0.05$), with a relationship between weight loss and storage time. During storage, there were weight losses in both sizes of scallops, with these losses being more pronounced in big scallops. Regarding the small scallops, until the ninth day of storage, there was always an increase in weight loss as the storage time increased. However, the weight losses began to be smaller after 9 days. Although not very accentuated, the temperature which led to greater weight loss was at 6 ºC and the one that took the lowest losses was at 3 ºC (Figure 19).

Regarding the big scallops, at 6 ºC the weight loss was more pronounced up to 3 days of storage (≈ 15%), stabilizing up to 6 days. After that, weight losses were almost proportional to storage time reaching 25% of total weight losses in 12 days. At 9 ºC, weight losses were almost proportional with the increase of the storage days, also at 25% in the end. At 3 °C, the scallops were losing more weight as the time of storage increased, reducing that loss from 20% to approximately 15% from the day 9 to the day 12 day (Figure 19).

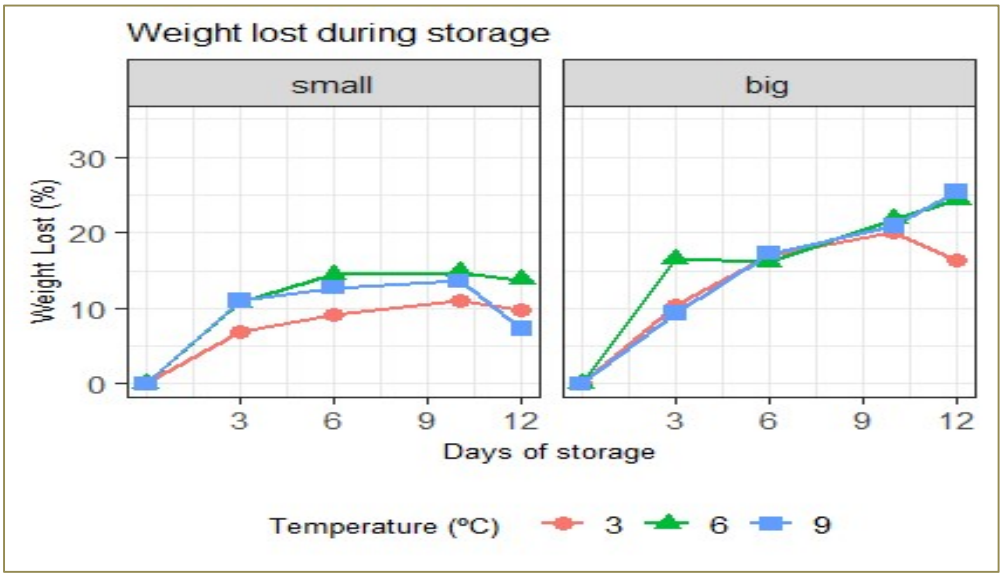

Figure 19: The weight lost (%) of the scallops during the storage at different temperatures (3, 6, and 9 ºC).

3.2.2 Survival Rate

After checking the survival of the scallops during storage, it was found that the number of survivors was inversely proportional to the storage time and temperature increasement. This relationship was statistically significant ($p < 0.05$). The temperature at which the survival was the highest was at 3 ºC. Thus, at 3 ºC died 50% of scallops until 12 days of storage (15 small scallops and 13 big scallops). However, the highest number of deaths occurred from the day 7. Therefore, it can be said that if the quality only depended on the survival, the scallops could be stored up to 7 days at 3 ºC, since only 3 scallops died in one week of storage. At 6 ºC, there is a difference between small and big scallops. While only 4 small scallops survived during the storage at this temperature, 6 big scallops survived. At 9 ºC, there were only 16 % of survivors (5 scallops small and 4 big scallops). The only surviving scallops at 6 and 9 °C were those stored until 3 days of storage. On the other days, there was 100% of mortality at both temperatures (Figure 20). Both temperature and storage days were significant ($p < 0.05$).

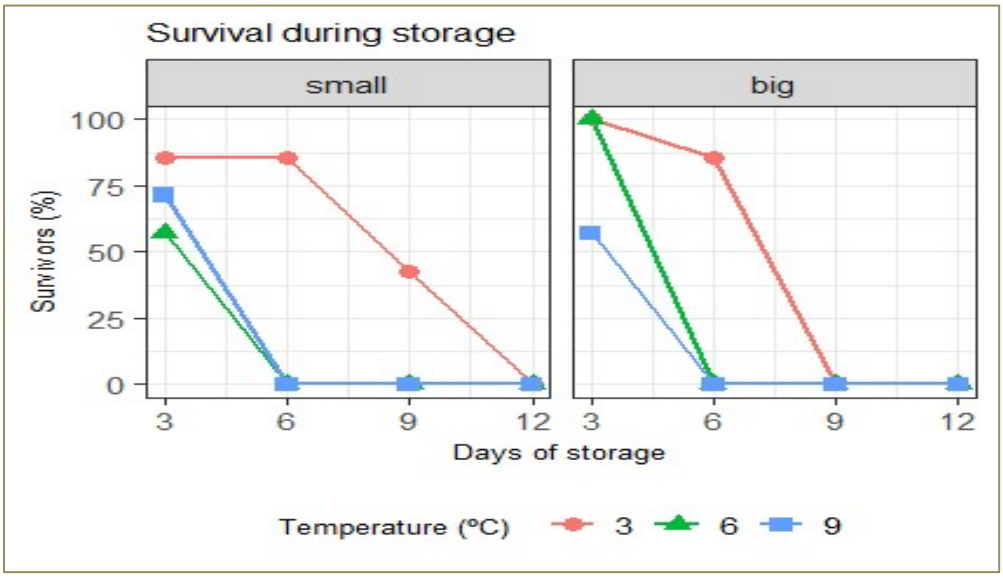

Figure 20: Survival of the scallops during storage at 3, 6, and 9 ºC. Each sample had a total of 7 scallops.

3.2.3 Sensory assessment

In total, 10 parameters were evaluated by the panel during the sensory test sessions. The results showed that there were certain deviations in sensory parameters at different temperatures during the different days.

Sulfhydric, putrid: This parameter is considered undesirable in a quality scallop and therefore is a negative parameter because when present, it can lead to a rejection of the product. During three days of storage, its presence was absent or very weak (intensity level = 1) in all seasons and at all temperatures. When the number of storage days increased, there was a strong increase in intensity. Thus, in March, the presence of this parameter exceeded the level (medium intensity) at 6 days and at 9 days at the maximum level: very intense (intensity level = 5) at 6ºC and at 9ºC. In June, at 3 and 6 ° C, the maximum intensity reached only level 4 (intense). There were differences in the level of intensity in the different sizes of scallops. While in March, the big scallops showed medium intensity (intensity level = 3) of Sulfhydric, putrid in 6 days of storage at 3ºC, the small oysters were almost absent (intensity level < 1). In August, the small scallops after 6 days at 6ºC of storage were evaluated with level 4 (intense) while the big scallops

were evaluated with level 2 (weak). The storage at 3 ºC registered the lowest level of intensity of this parameter, reaching the medium level only in March and June in the two sizes of the scallops. With $p < 0.05$, this parameter was statistically significant. The differences in intensity of Sulfhydric, putrid between temperatures and storage time were significant where $p < 0.05$ (Figure 21).

Ammonia: this is another parameter that indicates a scallop of poor quality, presenting values of intensity very similar to the previous parameter, with only a few variations (Figure 21). It was statistically significant ($p < 0.05$).

Sour: this negative parameter when present is associated with poor quality scallops. Once again, the intensity levels of this parameter obtained in the evaluation of the quality of the scallops were very similar to Sulfhydric, putrid and Ammonia, being the temperature of 3 ºC that showed lower values. As the parameters above, this was almost directly proportional with the increasing storage temperatures and increasing storage time. Also, the $p < 0.05$ shows that this parameter was statistically significant (Figure 21).

Musty: this attribute is also negative and therefore a scallop should not have high levels of intensity of this parameter. As the previous indicator, it was also statistically significant ($p < 0.05$). The storage temperature of 3 °C continues show the lowest intensity levels, increasing with the increasing days of storage. In March at 9 ºC, there was a huge increase in intensity from 3 days to 6 days of storage (from intensity level = 0 - absent to intensity level = 5 - very intense) but strangely, with the small oysters, the level of intensity decreases for the medium level (intensity level = 3) at 9 days of storage. In June, this parameter was always evaluated as absent (intensity level = 0) at all temperatures during all storage days (Figure 21).

Boiled milk, seaweed: It is one of the parameters that are considered positive when present in the scallops and therefore it is expected that the levels of intensity are high to obtain a quality scallop. In this study, this parameter never exceeded level 2 of intensity (intensity level = weak) and intensity levels decreased as the days of storage increased. With $p < 0.05$, this parameter was statistically significant. In relation to the big scallops, there was clearly a negative influence in the levels of intensity with the increase of the temperature and with the increase of the days of storage. However, there was an exception in August, since intensity levels were

higher at 6 days than at 3 days of storage. In relation to the small scallops, the time/temperature relationship was not so uniform, but the variability between the intensity levels was very small (Figure 21).

Fresh, seawater: considered as a positive parameter, the aroma of Fresh, sea water is very desired in a quality scallop. This parameter was statistically significant in this study ($p < 0.05$), where again the temperature of 3 °C had higher levels of intensity. Except for the small scallops in June, there was almost always a decrease in the intensity of this parameter as storage time and temperature increased. In June, there was variability in relation to the size of the scallops. At 3 ºC, the small scallops showed almost absent intensity levels (intensity level ≈0) whereas in the case of big scallops the intensity was almost medium (intensity level = 3) (Figure 21).

Slight blackening: this parameter is inconvenient in terms of quality and therefore, should have low levels of intensity. Thus, it is considered a negative parameter. The intensity of this attribute increased with increasing temperature and storage time. In March, it was evaluated as intense (intensity level = 4) at 9 °C with the big scallops and very weak (intensity level = 1) with small scallops. This parameter was significant in quality of scallops ($p < 0.05$) (Figure 21).

Brownish color: as the previous parameter, the big scallops stored in March got more intensity from Brownish yellowish color than the small scallops. However, in August, small scallops stored at 6 °C had a higher intensity of this attribute, this intensity being much lower after 3 days (intensity level was absent) than after 6 days of storage (intensity level was intense). This parameter was also statistically significant ($p < 0.05$) (Figure 21).

Slimy: This is one of the characteristics of scallops in a state of deterioration. Being a negative attribute, the intensity levels should be low in fresh scallops. With few exceptions, throughout the storage, the intensity of this parameter increased in both sizes at all temperatures. The highest intensity was reached in March with the big scallops at 6 and 9 °C after 12 days of storage. For the remaining storage time, the intensity only reached the medium level at the end of the storage time. This attribute was statistically significant ($p < 0.05$) (Figure 21).

Bright surface: A scallop considered fresh presents a bright surface and therefore, this parameter is considered positive. Since the p was < 0.05, this parameter was statistically

significant in quality of scallops. In this study, the intensity of this parameter was almost inversely proportional to the increase in temperature and the increase in storage time, except for two exceptions: in March, at 6 ºC in small scallops and at 9 ºC in big scallops there was an increase in intensity at from the day 6 (Figure 21).

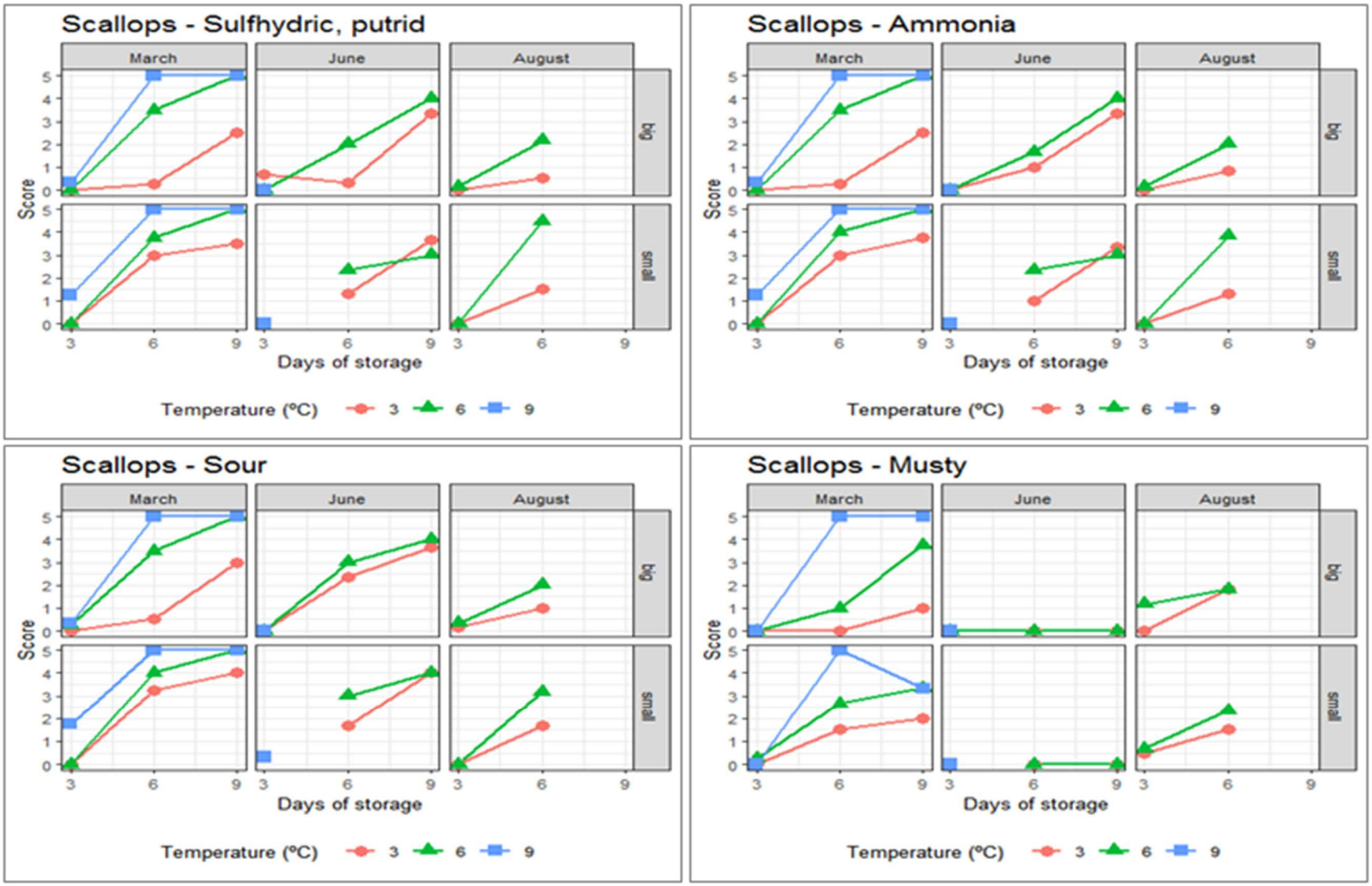

Figure 21: Average of the sensory evaluation of scallops during the storage, at different temperatures (3, 6, and 9 ºC). All the characteristics of the samples were evaluated by the participants with a level of the intensity ranged from level 0 to level 5 (0 - absent, 1 – very weak, 2 – weak, 3 – medium, 4 – intense, 5 – very intense).

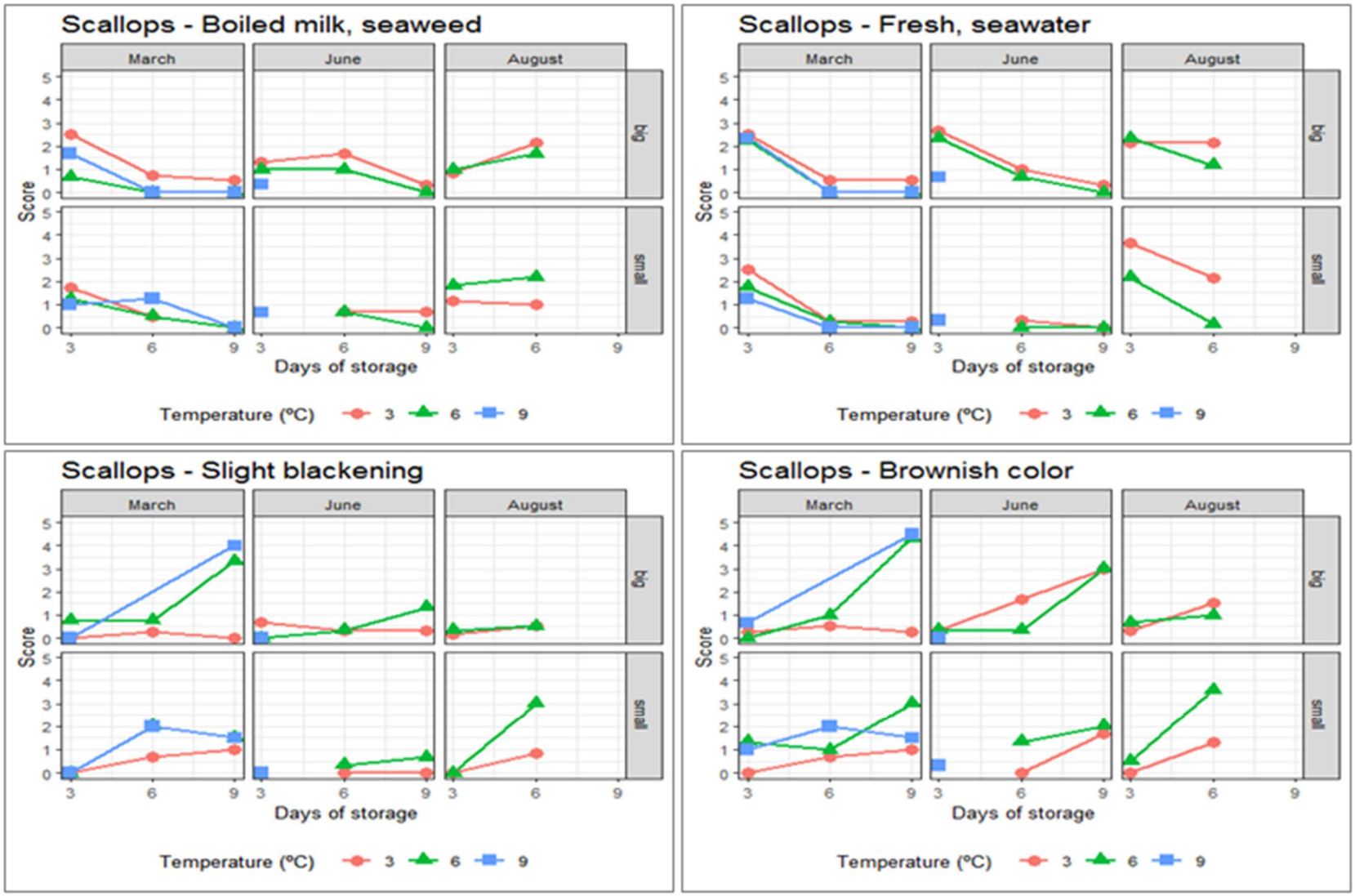

Figure 21: cont.

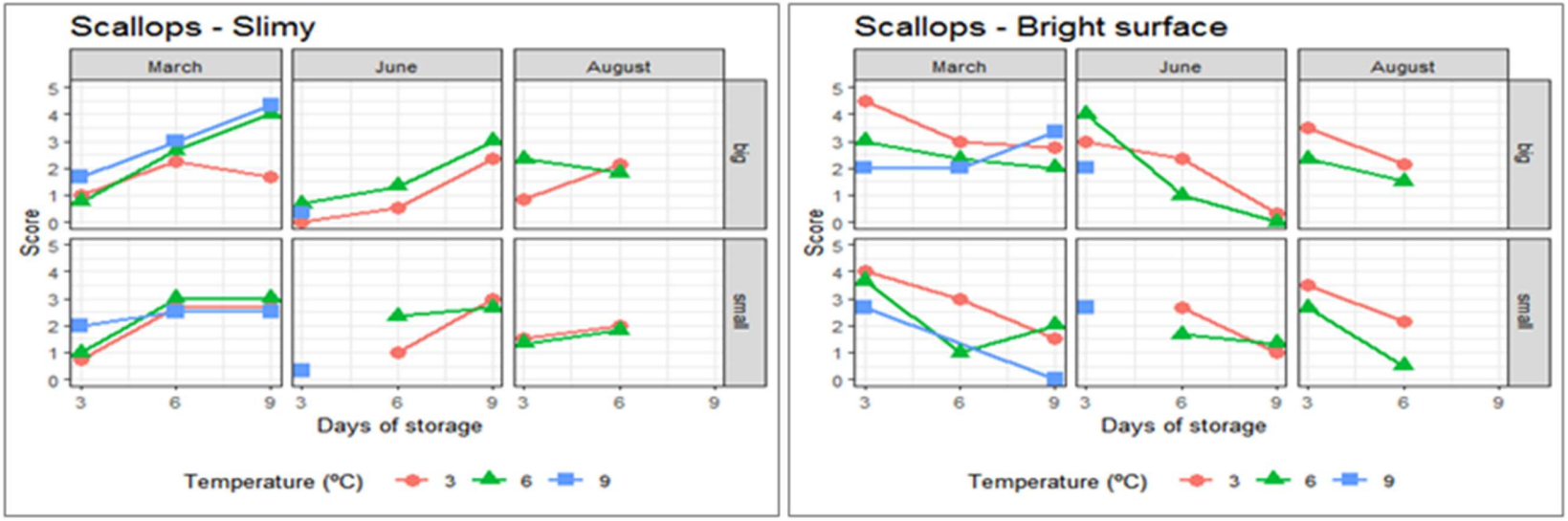

Figure 21: cont.

Scallops with a new cover: Replacing the cover of newsprint with a cover intended to come into contact with food was done in order to check whether there was an increase in the quality of the scallops with the new coverage. However, after the statistical analysis, it was found that there were not variability between the scallops with the new coverage and the big scallops stored in the same month (Figure 22). The $p > 0.05$ showed that it was not statistically significant.

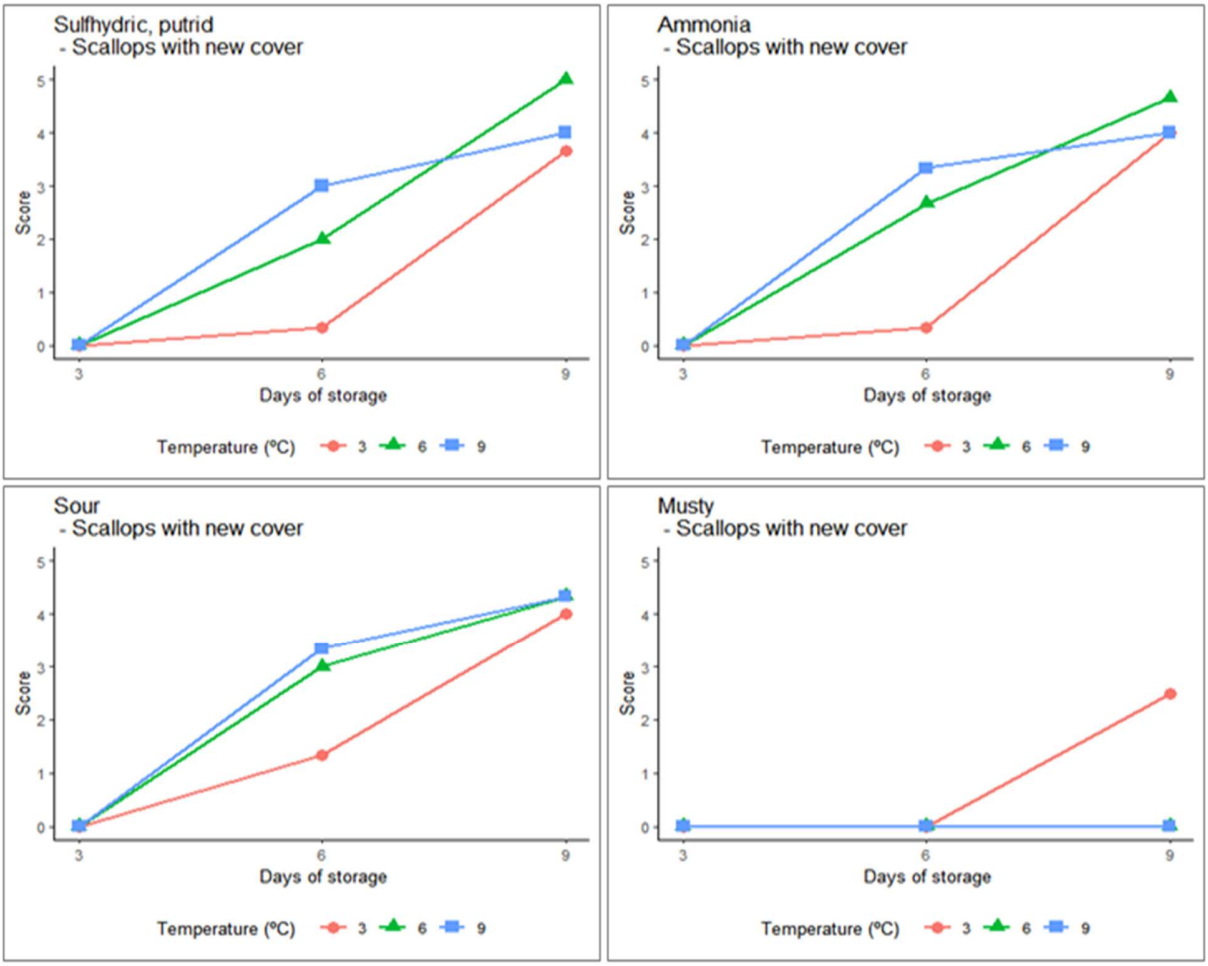

Figure 22: Average of the sensory evaluation of scallops during the storage, at different temperatures (3, 6, and 9 ºC). The scallops were placed in a box with a wet cover destinated to be in contact with food. All the characteristics of the samples were evaluated by the participants with a level of the intensity ranged from the level 0 to the level 5 (0 - absent, 1 – very weak, 2 – weak, 3 – medium, 4 – intense, 5 – very intense).

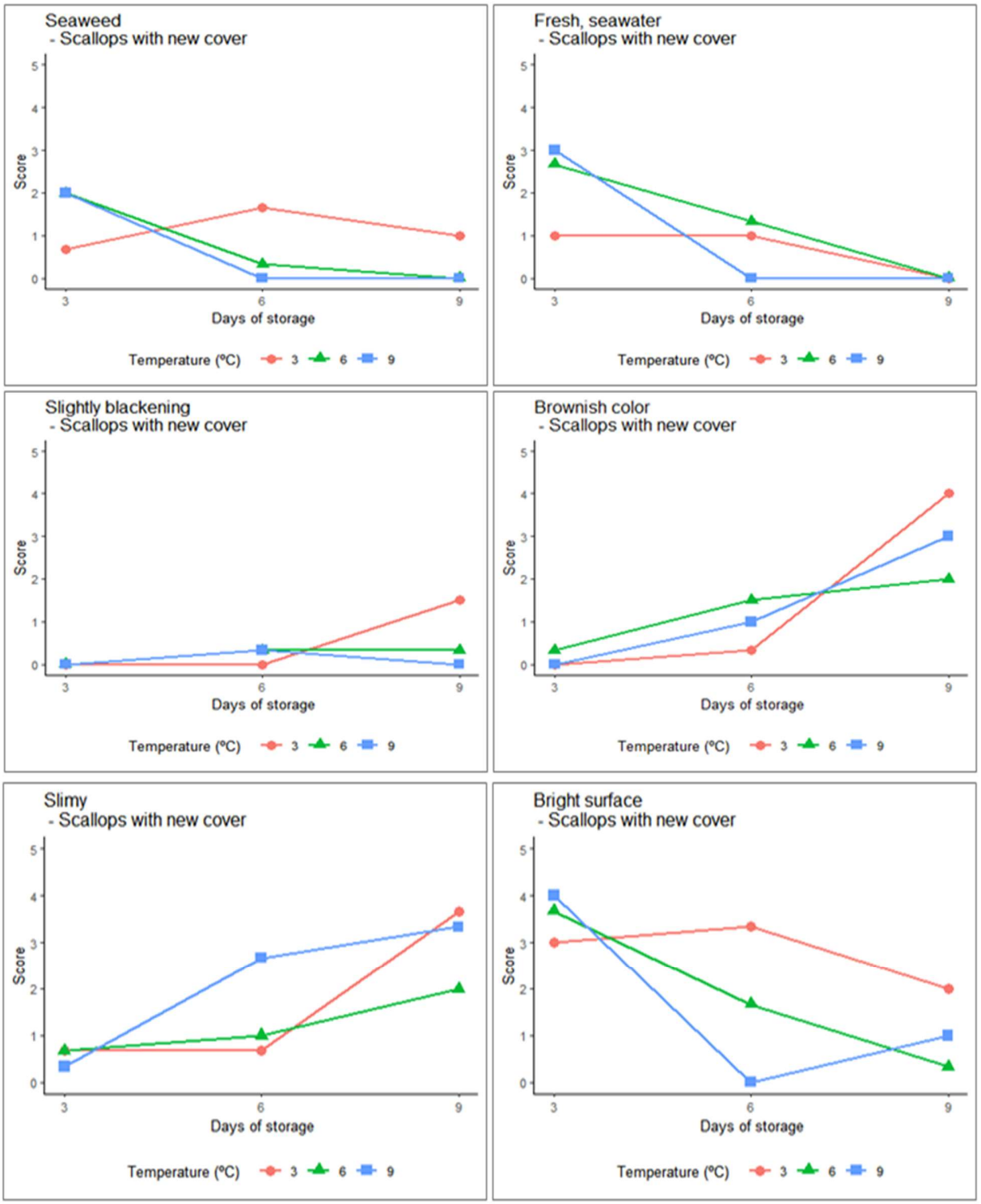

Figure 22: cont.

Scallops with diaper: The placement of an absorbent into the boxes under the scallops was aimed to absorb liquids released by the animals during storage, thereby maintaining the initial quality as long as possible. However, after obtaining the results of the sensory evaluation, it was checked that differences between the scallops with the absorbent and the big scallops (with normal package) were not statistically significant ($p > 0.05$) (Figure 23).

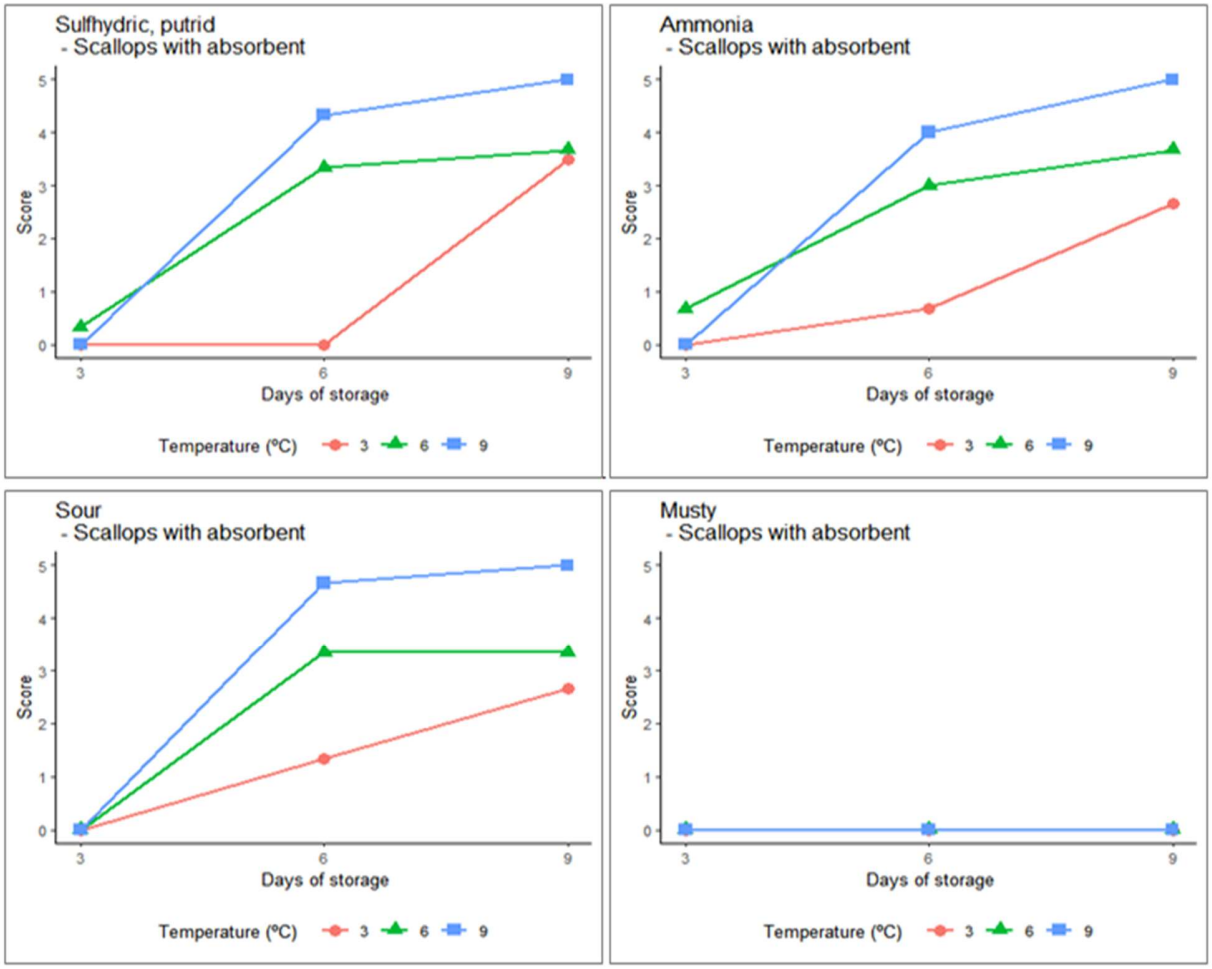

Figure 23: Average of the sensory evaluation of scallops during the storage, at different temperatures (3, 6, and 9 ºC). The scallops were placed in a box with an absorbent pad in order to absorb the liquids released from the animals. All the characteristics of the samples were evaluated by the participants with a level of the intensity ranged from the level 0 to the level 5 (0 - absent, 1 – very weak, 2 – weak, 3 – medium, 4 – intense, 5 – very intense).

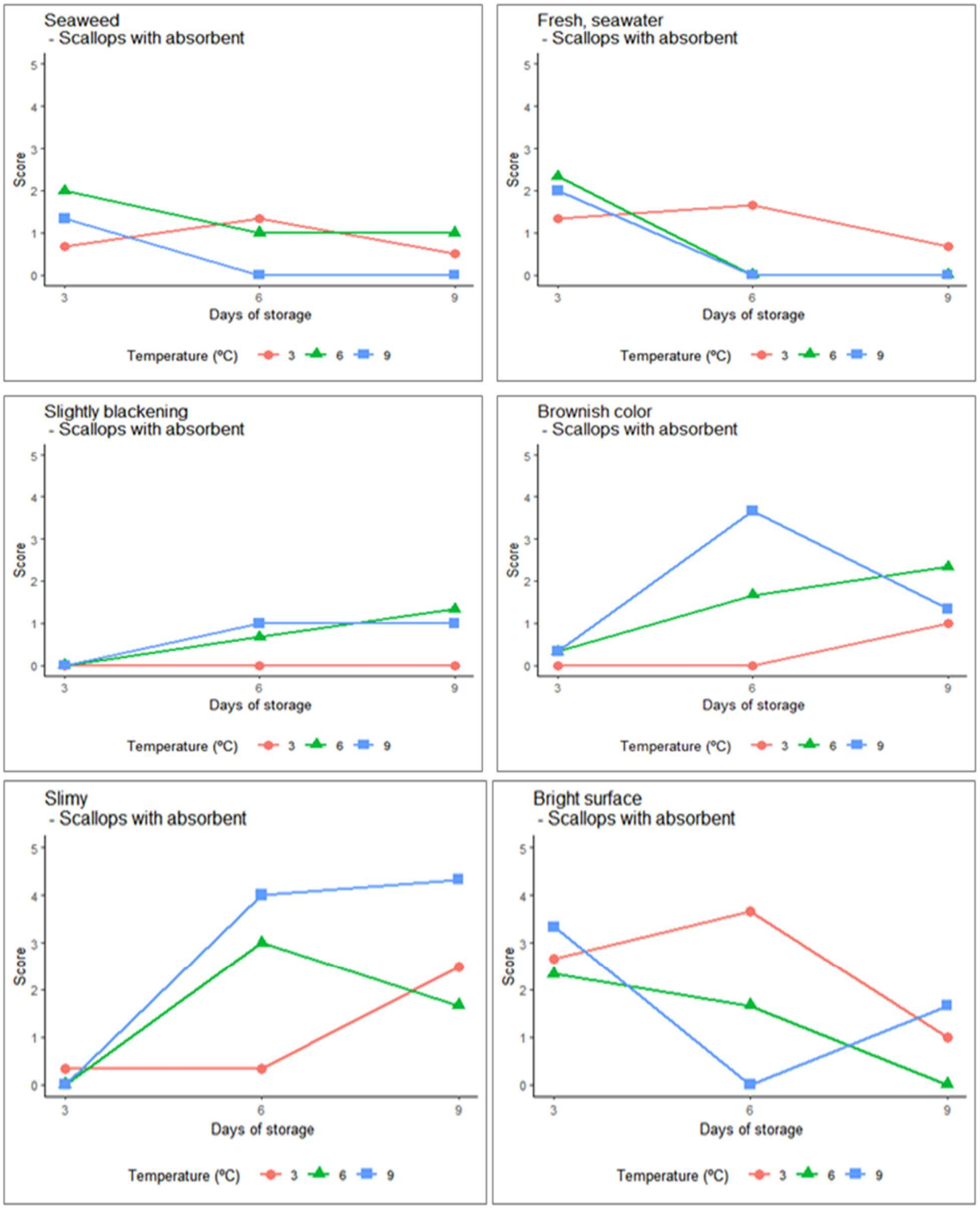

Figure 23: cont.

3.2.4 Acceptance of scallops

During the sensory evaluation, the participants were asked if the evaluated scallop samples were in good quality to be eaten. The acceptability of the scallops by the participants was practically inversely proportional to the days of storage (Figure 24). For three days of storage, the scallops had an acceptability above 75% in all seasons. When increasing to 6 days storage, the acceptability of this bivalve decreases, in some cases abruptly, to values of total rejection of the product (such as scallops stored at 9 °C in both sizes and small scallops stored at 6 °C in March and June). By increasing to the maximum storage time (9 days), there was a total rejection of the scallops by all participants. The factor "storage time" was significant ($p < 0.05$). The type of scallops (big scallops, small scallops, with a new cover and with the absorbent) was not significant ($p > 0.05$).

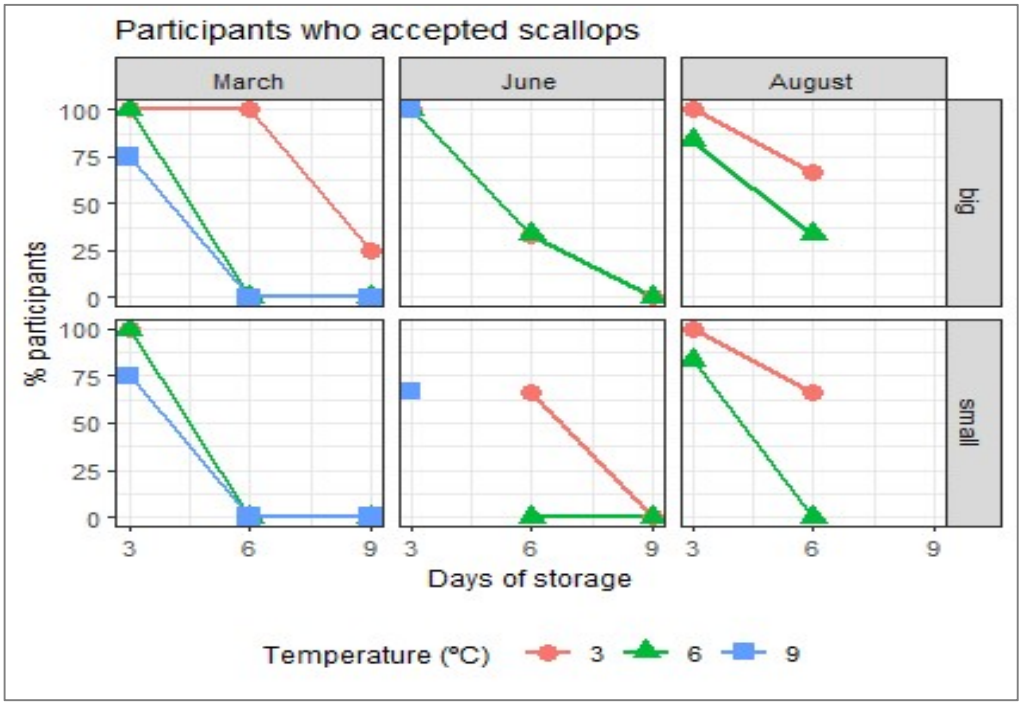

Figure 24: Acceptance of scallops by the participants**.** The number of participants who accepted the small scallops and big scallops is in percentage (%). In March, were carried out samples in 3, 6 and 9 days of storage at the three temperatures. In June, small scallops were stored only 6 and 9 days at 3 and 6 °C, and only 3 days at 9 °C. Big scallops were stored at 3, 6, and 9 days at 3 and 6 °C and in only 3 days at 9 °C. Both scallops with a new cover and those with absorbent were stored at 3, 6, and 9 days at all three temperatures. In August were only performed samples at 3 and 6 days of storage at 3 and 6 °C.

3.2.5 Quality level

When all parameters mentioned above are met, a method of evaluating the quality of the scallops during storage at a given time and at a given temperature can be obtained, with an overview of quality at three seasons. The graph below is the result of an average proportion of the intensity levels of the positive and negative parameters in order to achieve a quality determination of the scallops studied in this paper (Figure 25). The level "0" is the lowest level of quality. The levels below "2", means that the scallops have poor quality and should be rejected. The levels between "2" and "3" are the medium level while the levels above "4" are the best quality. As can be verified, the quality of the scallops is almost inversely proportional to the increase in storage time and the increase in temperature. The maximum level reached was level 4 and the minimum level was 0.5. The graph shows clearly that the temperature that reached the highest level of quality was 3 ºC, in all seasons (with levels never below 2), followed by the temperature of 6 ºC and lastly the temperature of 9 ºC. With regard to storage days, it is very noticeable that the lower the storage time, the higher the quality.

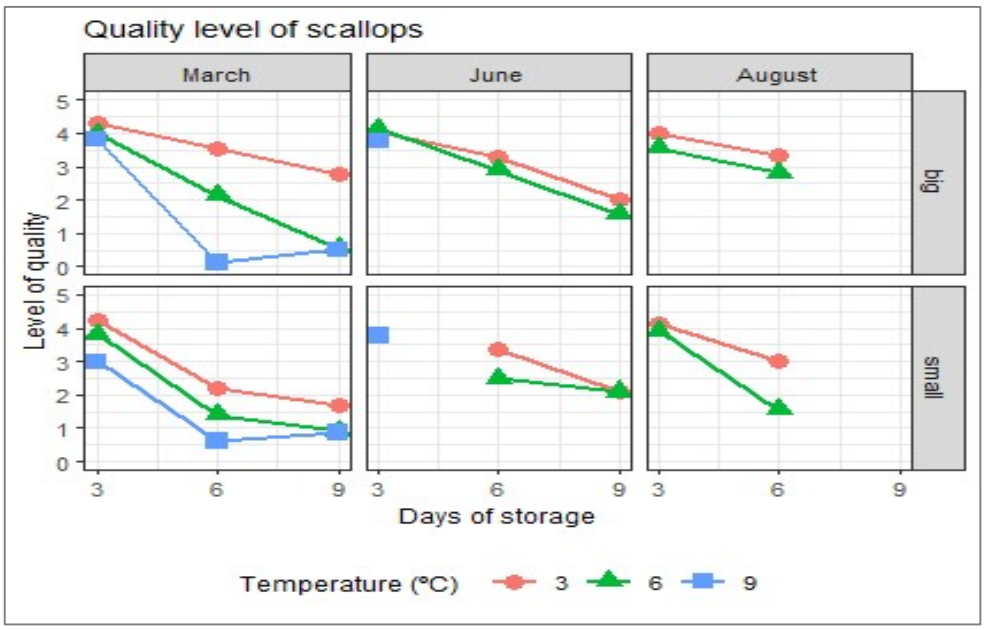

Figure 25: A quality overview of all scallops studied in this paper. The graph shows a ratio between negative and positive parameters, representing an overview of the quality of all scallops during storage at 3, 6 and, 9 °C, up to 9 days in three different seasons (March, June and August). The level "0" is the lower level of quality. The level between "2" and "3" is the medium level while the levels above "4" are the best quality.

Thus, storage for 3 days had the quality level 4 while at 6 days, the quality level ranged between 0 and 3. However, on the day 9 day of storage, the scallops never reached levels > 2. The time of the year when scallops were harvested was significant in the final quality of scallops ($p < 0.05$). The differences between March and August, and between June and August were significant ($p < 0.05$) while June and August were not significant ($p > 0.05$). The differences between days were significant ($p < 0.05$). Regarding the size of the scallops, the influence of this factor was not statistically significant in the final quality ($p > 0.05$).

3.3. Clams

3.3.1 Weight assessment

Differences in weight loss were statistically significant ($p < 0.05$). In the first 6 days of storage, weight losses were always less than 10%, and there was no great variability at the three temperatures. However, from the day 6, there were considerable weight losses reaching almost 30% by the day 12. It was at 9 ºC that was verified a higher weight loss in the first 6 days. Between 6 to 9 days of storage, the higher weigh losses were verified at 6 °C and after that period it was from 3 °C (Figure 26). The results showed that there were differences in the time of the storage ($p < 0.05$).

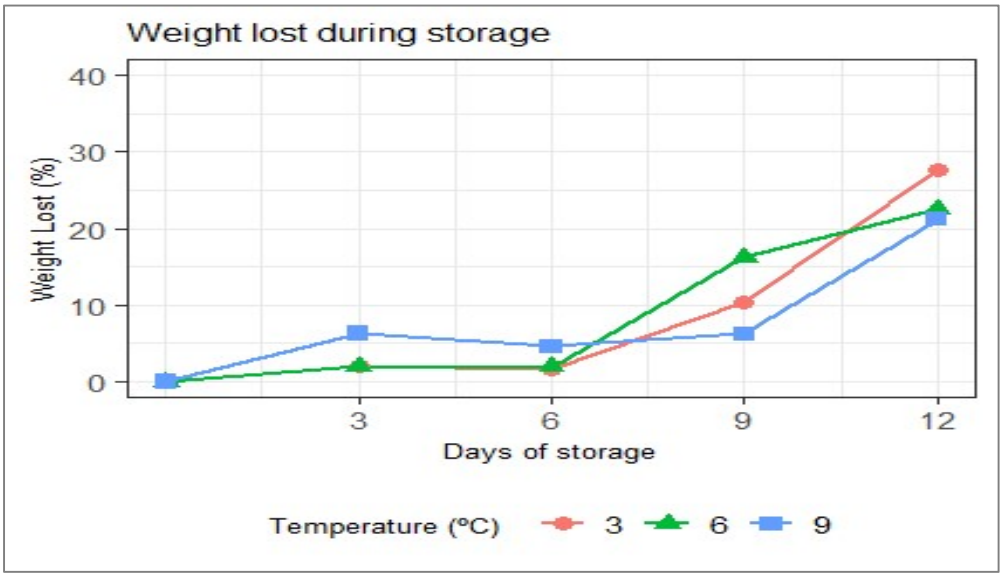

Figure 26: The weight lost (%) of the clams during the storage at different temperatures (3, 6, and 9 ºC). Differences in weight lost were statistically significant ($p < 0.05$).

3.3.2 Survival Rate

There were 5 deaths in 72 clams (Table 5). The temperature which there were more deaths was at 9 ºC and while at 6 ºC was any death. After 3 days, it was registered two deaths in 7 days, one death in 10 days and 2 deaths in 12 days of storage. Differences in survival were not statistically significant ($p > 0.05$).

Table 5: Number of clams which survived during the storage at different temperatures (3, 6 and 9 ºC). Each sample had a total of 6 clams. During the storage until 12 days, there were 5 deaths of 72 clams.

	Clams			
Temperature / Days	3 ºC	6 ºC	9 ºC	Total (survivors/total clams)
3 days	6	6	6	18/18
7 days	5	6	5	16/18
10 days	6	6	5	17/18
12 days	5	6	5	16/18
Total (survivors/total clams)	22/24	24/24	21/24	72

3.3.3 Sensory assessment

Fresh, algal, seawater: this is a positive parameter since it defines a clam with good quality and so the higher the level of intensity, the better the quality. In this study, the intensity level of this parameter was weak and the medium (between the intensity level 2 and the intensity level 3) in the two months (Figure 27). The intensity level was lower in the clams stored at 3 ºC than in the other temperatures in March after 12 days. In June, the variability was very small. The differences in Fresh, algal, seawater were not statistically significant ($p > 0.05$).

Sour: this parameter is considered negative when present in the clams. In the presented samples, the Sour was never evaluated with the level of intensity above absent/very weak. This parameter was not statistically significant ($p > 0.05$) (Figure 27).

Ammonia: like the previous parameter, the presence of ammonia is undesirable in the quality of clams. The results of the evaluation of this parameter showed that ammonia levels were absent or very weak in March and June for all three temperatures. The differences in Ammonia in quality of clams were not significant ($p > 0.05$) (Figure 27).

Sulfhydric, putrid: the levels of this parameter are very similar to the Ammonia levels, that is, they were always below level 1 (very weak). It is a very unpleasant smell and the levels of intensity have should be low. Sulfhydric, putrid was not significant in quality of clams ($p > 0.05$) (Figure 27).

Bright surface: this parameter is positive and so, the intensity level should be high. In this study, the levels of the Bright surface were between medium to intense (intensity level between 3 and 4) in March and June. This parameter was not statistically significant ($p > 0.05$) (Figure 27).

Cream color: the intensity of this parameter was evaluated as weak to medium (intensity level between 2 and 3) in March and in June for all temperatures of storage, except at 6 ºC in June which was evaluated with the level of intense (Figure 27). This parameter was not statistically significant ($p > 0.05$).

Color white: in March, the intensity of this parameter was between weak and medium at all temperatures (intensity level between 2 and 3). However, in June there was a great variability since the clams stored at 3 ºC presented Color white with a very weak intensity level whereas at 6 and 9 ºC the intensity level was intense (intensity level = 4) (Figure 27). Yet, this parameter was not statistically significant ($p > 0.05$).

Brownish color: in March, the intensity of this attribute was around level 3 (medium) at all temperatures. However, in June there was a great variability since the clams stored at 3 and 6 ºC presented Brownish color with a very weak intensity level whereas at 9 ºC the intensity level was absent (intensity level = 0) (Figure 27). The differences of Brownish color in quality were statistically significant ($p < 0.05$).

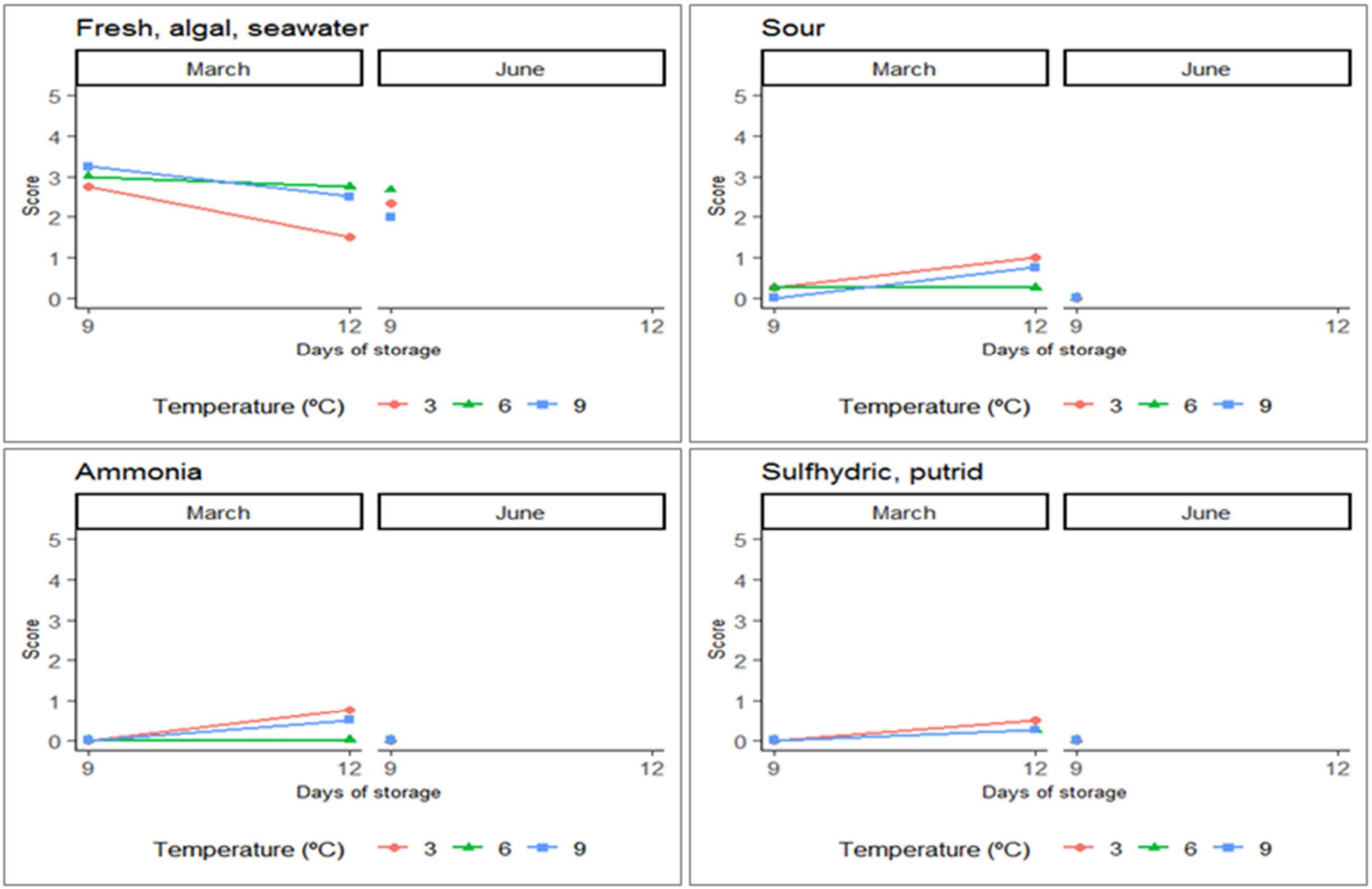

Figure 27: Average of the **s**ensory evaluation of clams during the storage, at different temperatures (3, 6, and 9 ºC). All the characteristics of the samples were evaluated by the participants with a level of the intensity ranged from the level 0 to the level 5 (0 - absent, 1 – very weak, 2 – weak, 3 – medium, 4 – intense, 5 – very intense).

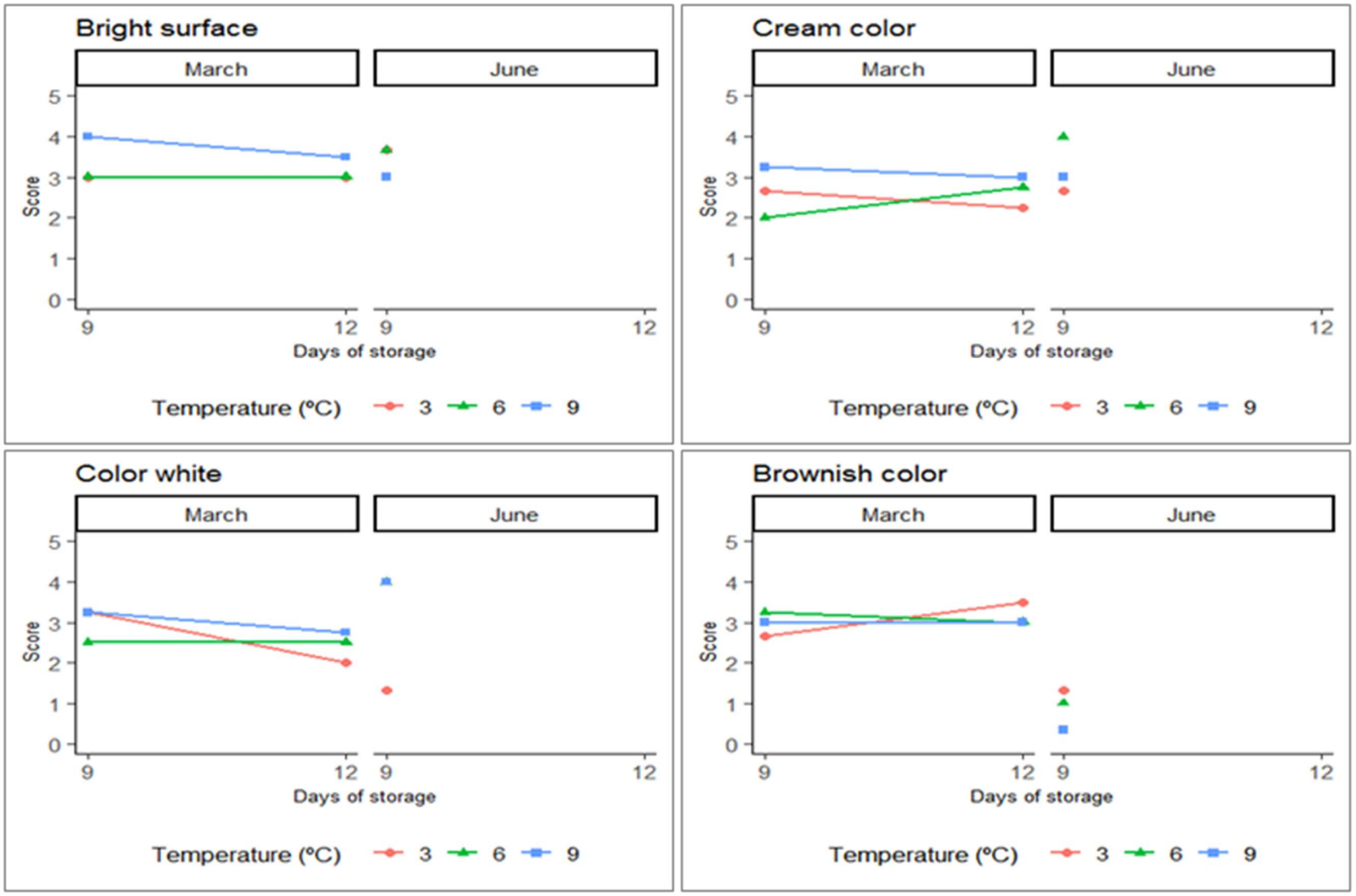

Figure 27: cont.

3.3.4 Acceptance of the clams

All participants found that the clams stored for 9 days at both 3 and 9 ºC were in good quality to be eaten. At 6 °C, 25% of the participants rejected the clams stored during the same time. On the other hand, clams stored 12 days at 6 °C (in March) were accepted by all participants whereas 3 °C clams were accepted by 75%, and the clams at 9 °C were only accepted by 50% of the participants (Figure 28). The differences in the acceptance of the clams between the storage times were significant ($p < 0.05$).

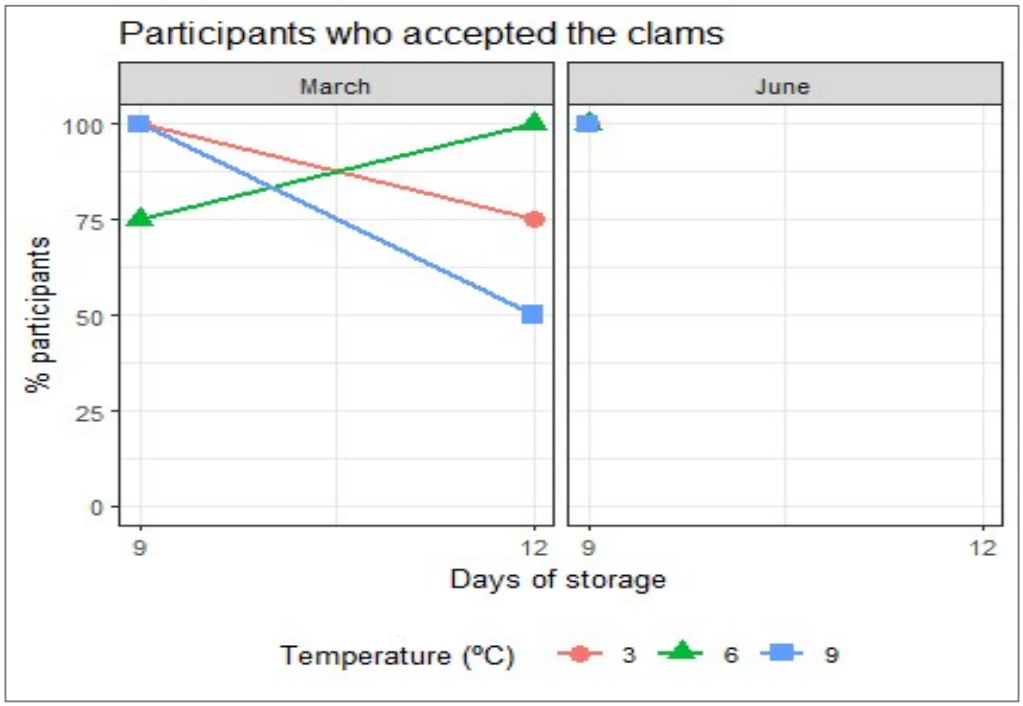

Figure 28: Acceptance of clams by the participants**.** The number of participants who accepted the clams is in percentage (%). In March, were carried out samples up to 12 days. In June, the clams were stored only for 9 days at the three temperatures.

3.3.5 Quality level

When all parameters mentioned above are met, a method of evaluating the quality of the clams during storage at a given time at a given temperature can be obtained, with an overview of quality in two seasons. The graph below is the result of an average proportion of the intensity levels of the positive and negative parameters in order to achieve a quality determination of the clams studied in this paper (Figure 29). The level "0" is the lowest level of quality. The levels below "2", means that the clams have poor quality and should be rejected. The levels between "2" and "3" are the medium level while the levels above "4" are the best quality.

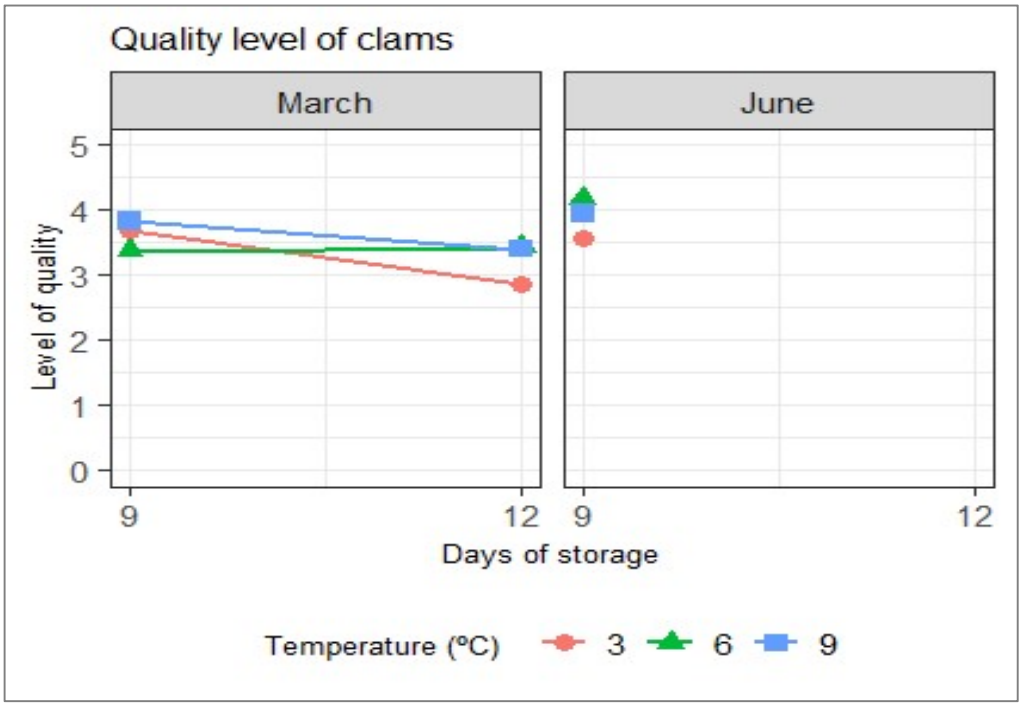

Figure 29: A quality overview of all clams studied in this paper. The graph shows a ratio between negative and positive parameters, representing an overview of the quality of all clams during storage at 3, 6 and, 9 °C, during 9 and 12 days in March, and 9 days in June. The level 0 is the lower level of quality. The level between 2 and 3 is the medium level while the levels above 4 are the best quality.

In March, the quality level was in level 3. Although was slight, the temperature which reaches the lowest level was at 3 ºC in 12 days of storage in March (level 2) and in 9 days in June (level 3). The quality level of clams was not statistically significant ($p > 0.05$).

In addition, it was noted in the whole experiment that clams had their valves great opened and the ones which were alive, closed after handling (Figure 30).

Figure 30: Clam with opened valves during the storage.

4. Discussion

Oysters

By converting and compiling the mean of all sensory evaluations of the positive and negative parameters, it was possible to obtain a quality gradient of all the samples. In this way, the level "0" is the lowest level of quality. The levels below "2" mean that the clams have poor quality and should be rejected. The levels between "2" and "3" are the medium level while the levels above "4" are the best quality. The quality levels of the oysters obtained in this study ranged between level 3 and 4. It means that almost all oysters had good quality. However, there were exceptions: with the small oysters stored for 6 days in March (6 and 9 ºC), and 9 days at 6 ºC in June; and with big oysters stored after 12 days at 9 ºC in March, after 9 days at 3 ºC in June and 3 and 6 days at 9 ºC in August. These oysters had medium quality. No oysters present in this study were considered to be of poor quality. In addition, the oysters' size was significant ($p < 0.05$) in the quality. Several authors have established shelf time limits for oysters, depending on different parameters. Cao et al. (2009) indicated a limit of 11 days, considering the sensorial evaluation and 10 days, considering the microbiological limits (107 CFU/g) for oysters stored at 5 ºC. On the other hand, Buzin et al. (2011) reported that after 22 days of storage at 3 °C and 100% moisture, oysters did not show significant organoleptic variations. In the case of raw mangrove oysters, Montanhini and Neto (2015) reported that they can be stored for 11 days if kept at temperatures between 10 and 15 ºC. The results of the present paper were consistent with those found by previous authors, since almost all samples were evaluated as good quality and few as medium quality.

Since oysters are preferably consumed in half shell, they should be presented to the consumer alive. Therefore, the survival of these bivalves after harvesting is very important for industry and linked to the final quality of the oysters. In this study, the survival rate of *O. edulis* was considered high with only 6 deaths in 168 animals stored at different temperatures until 12 days. Seaman (1991) reported that *Crassostrea gigas* survive for as long as 20 weeks out of water keeping the right temperature and humidity. However, although Aaraas et al. (2006) reported a high survival of oysters for 23 days of storage, the appearance and smell did not meet the quality standards. Therefore, they established a limit of 12 days, due to the growth of

deteriorating bacteria, which the limit is consistent with the present paper. Also, Cao et al. (2009) noted that the results of sensory analysis pointed to a shelf life of oysters bigger than the shelf life obtained by chemical analysis. These authors concluded that this phenomenon may be due to the fact that sensory evaluation is subjective and during the session of sensory assessment may not have been met organoleptic conditions for rejection of the product. It may also be due to the fact that the cold camouflaged the organoleptic characteristics of these bivalves, since oysters were stored at low temperatures. In the present study, there were some very strange values since some negative parameters were high in level of intensity and then decrease in the days remaining of storage. For instance, in the "Sour" parameter, the big oysters harvested in March, stored at temperatures of 6 and 9 ºC, obtained an intense level = 4 for 6 days, but this intensity decreased to very weak to absent (intensity level between 0 and 1) after the day 10. It can be due three explanations. First, this type of results may be due to the assessors' sense of smell could be fatigued due to the high number of sensory assessment day samples. The fact that only four advisors evaluated the quality of the product in March and only three in June led to the size of the panel being too small and therefore, unable to obtain a good rating. Second, the advisors' inexperience in sensory assessment sessions may have led to such strange results. It was discussed in the August session that there was some confusion among the participants in understanding the meaning of each parameter and their level of intensity, i.e., they were not sure which parameters meant inferior quality or which were the parameters that meant oysters of good quality. The third explanation could be due the intrinsic variability of each oyster. Aaraas et al. (2006) stated that there are individual variations between oysters and, since the batches placed on the market were of different quality, made it difficult to assess the freshness of these animals. In present paper, some evaluations of the negative parameters were more intense for 3 days than the 9 days of storage and the acceptability of the oysters by the assessors din not match with the sensorial analysis made by them. Therefore, it might be due to the oysters presented did not have quality, not due to the treatment post-harvest or the freshness, but due to the intrinsic qualities of the oysters. Duinker et al. (2008) concluded that both physiological processes and site of growth affected the taste and visual impression. The authors added that the most tasteful oysters were the those which were harvested in December and that the visual fullness, mineral taste and sweetness increase since September achieve the peak in the last month of the year. Also, it was reported by Beltrán-Lugo et al. (2006), Jiménez-Ruiz et al. (2015), Marquez Rios et al. (2007) and,

Montanhini and Neto (2015) that when living organisms change from common conditions to unusual environments due to, e.g. due to natural causes (such as seasonal changes) or due to human action, significant physiological changes in oysters may occur. Thus, in the new conditions, there is a decrease in glycogen levels, in total carbohydrate and ATP levels, causing biochemical changes in the muscle. Therefore, the occurrence of significant biochemical changes during harvesting, transport and storage can lead to a significant negative impact on final quality. This means that an oyster which has been stored for less time than the other oyster may have a lower quality due to the levels of stress which it has been subjected. In this way, due to these phenomena, in this study was hypothesized that the strange variability in the quality gotten in March and June was due to the type of oyster harvested, instead of being due to factors of sensory analysis. Although the oysters were of the same species and were supplied from the same company, *ScalMarin* traceability of the animals was not performed. As scuba divers deliver their product on site, the oysters are placed in a tank along with those which were delivered by other divers, in previous days. Therefore, the site, the harvest day, the time stored in tanks, and other factors may have varied in the same sample of oysters used for this study, thus causing great variability. In order to prevent this variability, in August was requested oysters of the same batch. It means that the oysters were delivered by the divers to the premises of the *ScalMarin* on the same day. However, even guaranteeing the same batch of oysters, the quality gotten in August compared to the quality gotten in both June and March was not statistically significant ($p > 0.05$).

Scallops

Considering the conversion and compilation of the mean of all sensory evaluations of the positive and negative parameters of the quality product, the scallops with lowest levels were those which stored at 9 ºC. The storage limit for scallops stored at 6 and 9 °C should be set for a maximum of 3 days, while those stored at 3 °C can be stored for up to 7 days. In this way, the scallops that obtained the best level of quality were those that were stored at 3 ºC. The finds were not in agreement with Ruiz-Capillas et al. (2001) neither Maxwell-Miller et al. (1982). The first study, the authors concluded that the maximum possible shelf-life for thawed king scallop meats was 9 days. The second study, the authors which studied Purple-hinge rock scallops (*Hinnites*

multirugosus), concluded that there were no significant differences in most sensory attributes or in acceptability of cooked scallops during 14 days of chill storage, and at no time in the storage period did the average ratings fall into the category of the rejection. The results of the present study show that the evaluation of the scallops stored for 9 days got low levels of quality at all the temperatures. These results can be explained with the survival of these bivalves during storage. From the day 3, there was 100% mortality of scallops stored at 6 and 9 °C whereas at 3 °C, this rate was only reached from day 7. It means that as the temperature and storage time increases, mortality also increases. These results are according with the results of the Duncan (1993) who proved that the mortality increase with the rise of emersion temperature. The author stated that high temperatures are responsible for decreasing energy levels, by the accumulation of toxins, leading to a decrease in pH. Bacterial growth is also faster, leading the faster cell deterioration. Therefore, increasing the temperature increases the mortality and turn decreases the quality of the scallops. Before performing the experiments, there was the idea that the scallops died at temperatures below 4 °C but which nevertheless could keep the quality thereof for a while, even if not living. However, this idea fell apart because the mortality of scallops at 3 °C was very low until 7 days of storage and after that day, the quality level decrease to unacceptable values.

Parameters considered being positives in the quality of scallops, rarely presented high levels of intensity (intense or very intense) in this study. This may be due to the fact that the negative parameters, when present, are very senses even at low concentrations, as they are undesirable.

According to Christophersen et al. (2008), adult scallops have long tolerated exposure to air than juvenile scallops. However, in the present study, this variation was not verified since the differences of scallops size were not a statistic significant ($p > 0.05$) neither in survival ($p > 0.05$).

The intensity levels of the scallop parameters stored with both the new cover and the absorbent were very similar to the intensity levels of the big scallops. The variance within the scallop type group (between small scallops, big scallops, scallops with absorbent and scallops with the new cover) was not statistically significant since the *p-values* were > 0.05. According to Otoni et al. (2016), the absorbent pads are one of the most successful applications of active food packaging systems since can retard the growth of spoilage microorganisms. Although in the

present study, the scallops with an absorbent pad were not significant different through evaluation of the quality, a decrease of the unpleasant smell was felt when the boxes were opened by the researcher. This suggests that there may have been a decrease in ammonia concentrations and a lower development of microorganisms relative to other stored scallops without any type of absorbent.

Other factors which were studied in this paper were the influence of the seasonality of the scallops in relation to the final quality. Strohmeier et al. (2000) suggested that the energy allocation of the scallops can be divided into two parts in the year: since winter to early summer, is for reproductive growth and the other part, since late summer to autumn, is for somatic growth and storage. The conclusions of Christophersen et al. (2008) are in agreement with these authors, since they concluded that scallops showed higher tolerance to transport conditions during the cold-water season. Also, in the present paper, the quality of scallops varied with the season of the harvest ($p < 0.05$), which fit with the results of the previous authors. Comparing the seasons, March was different from June and August ($p < 0.05$) while June was not different from August ($p > 0.05$). March (winter) is precisely the time that scallops have fewer carbohydrate reserves due to hibernation and due to the mobilization of energy for the development of the gonads. The beginning of autumn is the time of year when there are more energy reserves in the scallops (Duncan et al., 2016; Idler et al., 1964; Seafish, 2013). The sexual variation and therefore, the biochemical variability of the scallops during the different seasons, can explain the variability of the quality of the scallops in this study.

The presence of living organisms (marine animals and plants) on the scallops was verified several times while weighing and preparing the boxes for storage. The presence of other living organisms in the scallops may have negatively influenced the final quality since they may have contributed to the further development of undesirable microorganisms and chemicals. (Boulter, 1996) reported that scallops contaminated with grit and mud deteriorate faster. For better quality assurance, farms must routinely perform gentle cleaning operations on scallop shells in order to prevent foreign organisms from being present at the time of packaging.

Clams

There were only some deaths during the storage of clams and, the differences throughout the storage period at all three temperatures were not significant.

Regarding the quality of the clams, there were no significant differences between the two seasons nor different temperature and storage time. The parameters which are considered negative (Ammonia, Sulfhydric, and Sour) had low intensity levels in this study. The uniformity observed in the clams quality matches with the low mortality rate during storage. Strangely, regarding to the acceptance of the clams by the participants was different. In March, 25% of the participants rejected the clams stored at 6 ºC for 9 days but for 12 days nobody rejected. Also, half of the participants rejected the clams at 9 ºC for 12 days. These strange results may be due not to the intrinsic qualities of the clams but due to the appearance after shucking the samples. Some of the participants were masters in bivalves shucking. During the sensory assessment sessions, they referred that there were gotten some bivalves which were shucked improperly. Several studies (Altintzoglou and Heide, 2016; Grunert, 2005; Martinsdóttir et al., 2009; Mueller Loose et al., 2013) pointed that the appearance of seafood is one of the most important factors of consumer acceptance. The handling of the fish, made before being presented, will influence the perception of the quality of the consumer. While there are many methods to cut fish, it is the skill acquired with practice that will dictate end products with superior quality (Bykowski and Dutkiewicz, 1996).

In a study made by Sadok et al. (2004), was reported 50 % of mortality of in live, stored clam, *Tapes decussatus* for 16 days at 5 and 10 ºC. In another paper, Torres (2011) concluded that the clams stored for 3 days at 9 ° C were not fit for consumption. In the same study, the authors verified a mortality of 50% of these bivalves stored for 5 days at 4 °C. The results of the present study do not match to the results of these two studies since besides the mortality was very low, the clams stored at 9 days were evaluated with good quality levels. However, high survival of *Arctica islandica* in this paper are in agreement with Anacleto et al. (2013) who reported high survival in the transport for 9 days at 4ºC of the clams *Ruditapes philippinarum*.

During storage, a decrease in the weight of the clams was probably recorded by leakage. However, these fluid losses did not affect the final quality. According to Ali and Nakamura

(1999), this type of loss can facilitate gas exchange with the atmosphere since it exposes a large area of fluid from the mantle cavity to the air. In this study was checked that the clams had the valves open during storage. This phenome can be related to the clams' ability of exposure to the air. The previous authors showed a relationship between the differences in air breathing rates and the degree of shell opening. They demonstrated that the bivalves with an upper aperture degree of the shells during aerial exposure showed oxygen consumption rates also higher than those with semi-closed or closed shells.

5. Conclusions

After evaluating all the parameters obtained through sensory analysis, carried out by the participants, with this study it can be concluded in general, the oysters always presented good quality levels throughout the storage at different temperatures, and there was no evidence of seasonality since the differences of seasons in quality were not statistically different ($p > 0.05$). No oysters presented in this study were considered to be of poor quality. Therefore, it can be concluded that oysters can be stored until 12 days without losing qualities, at 3, 6 and 9 ºC. In this study, there was a strange variability concerning oyster quality results. This variability may have been due to the fact that there is variability among individuals from the same batch. Although in August it was ensured that the oysters supplied were harvested from the same day, there were no significant differences from the other seasons of the year through sensory analysis.

Regarding the scallops, after the analysis of all results, it can be concluded that there is a relationship between the survival and the quality. Also, the bad results of the quality matched with the mortality rate, concluding that the quality and survival during storage were related. Therefore, it can be stated that quality levels go down to unacceptable values for the consumer when the stored scallops die. In addition, the variability within the scallop type group (between small scallops, big scallops, scallops with absorbent and scallops with the new cover) was not statistically significant ($p > 0.05$). For further studies, it is suggested that other quality analyzes of the scallops stored in different packages should be carried out. The season of the year in which the scallops were harvested had a relationship with the final quality of the product ($p < 0.05$).

Regarding the clams, there were detected only few deaths during the storage and the differences during the whole storage time at the three temperatures were not significant. Thus, the survival of the clams was not affected during 12 days of storage at 3, 6 or 9 ºC. The clams, evaluated by the participants, presented good quality levels throughout the storage at the different temperatures, and there was no evidence of seasonality ($p > 0.05$). No clams presented in this study were considered to be of poor quality. Therefore, it can be concluded that clams can be stored until 12 days without losing qualities, at 3, 6 and 9 ºC. The high survival and the quality levels achieved after the evaluation of the clams suggest that this bivalve has a good resistance to anoxic stress.

In this study, some strange values were obtained, after the evaluation of bivalves. These results may have been due to some factors:

1) Due to the difficulty in obtaining available people with good knowledge of bivalves and able to evaluate the quality of these molluscs, the panel was sometimes small.

2) The sensorial evaluation had to be done with a large number of bivalves which could have led to the fatigue on the part of the participants, even with the implementation of procedures to avoid it.

3) Although the participants were very knowledgeable about the freshness of the bivalves and handled the bivalves almost daily, they may not have been able to express this knowledge through surveys.

4) Some participants were masters in the opening/shucking of bivalves. The fact that this manipulation was not carried out by a professional could have led the participants to reject the product, without however being rejected due to changes in quality due to storage.

5) The individual variability of bivalves due to the fact that they do not belong to the same batch may have affected the final quality assessment.

Therefore, for further studies, it is necessary to ensure that all participants understand the surveys well so that they can express in the questionnaires the variability of the presented samples. In addition, although often difficult, a panel with a larger number of participants is advised, as well as a smaller number of samples per session. In addition, quality control is advised through more subjective parameters such as chemical and microbiological parameters.

After analysis of all results it can be concluded that, living oysters and clams generally had good quality after dry storage for 12 days up to 9 °C, whereas the scallops were only of good quality for 7 days at 3 °C and 3 days for temperatures at 6 and 9 °C. The seasonality effect was significant associated in with scallops but not with oysters and clams.

6. References

Aaraas, R., Hernar, I.J., Vorre, A., Bergslien, H., Lunestad, B.T., Skeie, S., Slinde, E., Mortensen, S., 2006. Sensory, histological, and bacteriological changes in Flat oysters, *Ostrea edulis* L., during different storage conditions. Journal of Food Science 69, 205–210. https://doi.org/10.1111/j.1365-2621.2004.tb11006.x

Alasalvar, C., Taylor, T., 2002. Seafoods - Quality, Technology and Nutraceutical Applications.

Ali, F., Nakamura, K., 1999. Effect of temperature and relative humidity on the tolerance of the Japanese clam, *Ruditapes philippinarum* (Adams & Reeve), to air exposure. Aquaculture Research 30, 629–636. https://doi.org/10.1046/j.1365-2109.1999.00303.x

Altintzoglou, T., Heide, M., 2016. Fish quality and consumers: how do consumers' knowledge about and involvement in fish quality define factors that influence fish buying behavior? Journal of Aquatic Food Product Technology 25, 885–894. https://doi.org/10.1080/10498850.2014.964432

Anacleto, P., Maulvault, A.L., Barrento, S., Mendes, R., Nunes, M.L., Rosa, R., Marques, A., 2013. Physiological responses to depuration and transport of native and exotic clams at different temperatures. Aquaculture 408–409, 136–146. https://doi.org/10.1016/j.aquaculture.2013.05.035

Aníbal, J., Esteves, E., Rocha, C., 2011. Seasonal variations in gross biochemical composition, percent edibility, and condition index of the clam *Ruditapes decussatus* cultivated in the Ria Formosa (South Portugal). Journal of Shellfish Research 30, 17–23. https://doi.org/10.2983/035.030.0104

Aquamaps, 2016a. Reviewed distribution maps for *Ostrea edulis* (edible oyster), with modelled year 2100 native range map based on IPCC A2 emissions scenario [Online]. Available from: www.aquamaps.org (accessed 11.16.2018).

Aquamaps, 2016b. Distribution maps for *Arctica islandica* (Ocean qauhog), with modelled year 2100 native range map based on IPCC A2 emissions scenario [Online]. Aquamaps. Available from: www.aquamaps.org (accessed 11.16.2018).

Archer, M., 2010. Sensory assessment scoresheet for fish and shellfish.

Barrento, S., Keay, A., Lupatsch, I., 2013. Protocol on best practice handling and transportation of live mussels (Technical Report No. 243452). SME.

Bartelme, T.D., 2004. Short take: stress in fish, part I: What is stress in fish? Advanced Aquarist III.

Beltrán-Lugo, A.I., Maeda-Martinez, A., Pacheco-Aguilar, R., Nolasco, H., 2006. Seasonal variations in chemical, physical, textural, and microstructural properties of adductor muscles of Pacific lions-paw scallop (*Nodipecten subnodosus*). Aquaculture 258, 619–632.

Boulter, M., 1996. Scallop Quality - The effects of different handling practices and storage times, prior to processing, on scallop quality and investigation of quality assessment methods/indicators including D-Lactic acid (No. SR469). The Seafish Industry Authority.

Brand, A.R., Roberts, D., 1973. The cardiac responses of the scallop *Pecten maximus* (L.) to respiratory stress. Journal of Experimental Marine Biology and Ecology 13, 29–43. https://doi.org/10.1016/0022-0981(73)90044-0

Brayden, W.C., Noblet, C.L., Evans, K.S., Rickard, L., 2018. Consumer preferences for seafood attributes of wild-harvested and farm-raised products. Aquaculture Economics & Management 22, 362–382. https://doi.org/10.1080/13657305.2018.1449270

Frederiksen, M., 2002. Safety and quality issues in fish processing, in: Bremner, H.A. (Ed.), Quality Managment of of Stored Fish, Woodhead Publishing in Food Science and Technology. Taylor & Francis.

Buzin, F., Baudon, V., Cardinal, M., Barillé, L., Haure, J., 2011. Cold storage of Pacific oysters out of water: biometry, intervalval water and sensory assessment. International Journal of Food Science & Technology 46, 1775–1782. https://doi.org/10.1111/j.1365-2621.2011.02686.x

Bykowski, P., Dutkiewicz, D., 1996. Freshwater fish processing and equipment in small plants (FAO Fisheries Circular No. 905). FAO, Rome.

Caddy, J.F., 1973. Underwater observations on tracks of dredges and trawls and some effects of dredging on a scallop ground. Journal of the Fisheries Research Board of Canada 30, 173–180. https://doi.org/10.1139/f73-032

Campbell, M.J., Officer, R.A., Prosser, A.J., Lawrence, M.L., Drabsch, S.L., Courtney, A.J., 2010. Survival of graded scallops *Amusium balloti* in Queensland's (Australia) trawl fishery. Journal of Shellfish Research 29, 373–380. https://doi.org/10.2983/035.029.0213

Cao, R., Xue, C.-H., Liu, Q., Xue, Y., 2009. Microbiological, chemical, and sensory assessment of Pacific Oysters (*Crassostrea gigas*) stored at different temperatures. Czech Journal of Food Sciences 27, 102/108.

Cashmore, D., MacMillan, J.T., Learmouth, M., 1996. Methods of transportation for scallops (*Pecten maximus* L.) (No. 481). Seafish Industry Authority.

Chandrapavan, A., Kangas, M.I., Sporer, E.C., 2012. Seasonal, Spatial, and Postharvest Variability in the Survival of Repeatedly Discarded Saucer Scallops in Shark Bay, Western Australia. Journal of Shellfish Research 31, 1161–1171. https://doi.org/10.2983/035.031.0427

Chen, L., 2011. Sensory and Chemical Characteristics of Eastern Oysters (*Crassostrea virginica*). Virginia Polytechnic Institute and State University, Blacksburg, Virginia.

Cheney, D.P., 2010. Bivalve Shellfish Quality in the USA: From the Hatchery to the Consumer. Journal of the World Aquaculture Society 41, 192–206. https://doi.org/10.1111/j.1749-7345.2010.00348.x

Christophersen, G., Román, G., Gallagher, J., Magnesen, T., 2008. Post-transport recovery of cultured scallop (*Pecten maximus*) spat, juveniles and adults. Aquaculture International 16, 171–185. https://doi.org/10.1007/s10499-007-9135-9

Cooper, L., n.d. Using Fish and Shellfish Quality Assessment Methods.

Coton, M., Joffraud, J.J., Mekhtiche, L., Leroi, F., Coton, E., 2013. Biodiversity and dynamics of the bacterial community of packaged king scallop (*Pecten maximus*) meat during cold storage. Food Microbiology 35, 99–107. https://doi.org/10.1016/j.fm.2013.02.011

Cranford, P., Reid, G., Robinson, S., 2013. Open water integrated multi-trophic aquaculture: constraints on the effectiveness of mussels as an organic extractive component. Aquaculture Environment Interactions 4, 163–173. https://doi.org/10.3354/aei00081

Dridi, S., Romdhane, M.S., Elcafsi, M., 2007. Seasonal variation in weight and biochemical composition of the Pacific oyster, *Crassostrea gigas* in relation to the gametogenic cycle and environmental conditions of the Bizert lagoon, Tunisia. Aquaculture 263, 238–248. https://doi.org/10.1016/j.aquaculture.2006.10.028

Duinker, A., Mortensen, S., Nesse Okland, S., Bergtun, E., 2008. Visual and tasteful evaluation of oyster quality. Journal of Shellfish Research 27, 1004–1004.

Duncan, P.F., 1993. Post-harvest physiology of the scallop *Pecten maximus* (L.). (PhD Thesis). University of Glasgow, Glasgow.

Duncan, P.F., Brand, A.R., Strand, Ø., Foucher, E., 2016. The European scallop fisheries for *Pecten maximus*, *Aequipecten opercularis*, *Chlamys islandica*, and *Mimachlamys varia*, in: Developments in Aquaculture and Fisheries Science. Elsevier, pp. 781–858. https://doi.org/10.1016/B978-0-444-62710-0.00019-5

Encyclopedia Britannica, 2018. Oyster. Encyclopedia Britannica.

FAO, 2018. Species Fact Sheets *Arctica islandica* (Linnaeus, 1767) [Online]. Food and Agriculture Organization from United Nations. Available from: http://www.fao.org/fishery/species/3534/en (accessed 6.11.2018).

FAO, 2017a. Species Fact Sheets, *Ostrea edulis* (Linnaeus, 1758) [Online]. Food and Agriculture Organization of the United Nations. Available from: http://www.fao.org/fishery/culturedspecies/Ostrea_edulis/en (accessed 11.16.2017).

FAO, 2017b. Species Fact Sheets, *Pecten maximus* (Linnaeus, 1758) [Online]. Food and Agriculture Organization of the United Nations. Available from: http://www.fao.org/fishery/species/3516/en (accessed 11.16.2017).

Gage, S.D., Gorham, F.P., 1925. Self-purification of oysters during the hibernation. Am J Public Health (N Y) 15, 1057–1061.

Gercken, J., Schmidt, A., 2014. Current status of the European Oyster (*Ostrea edulis*) and possibilities for restoration in the German North sea. Federal Agency for Nature Conservation, Germany.

Gonçalves, A., Pedro, S., Duarte, A., Nunes, M.L., 2009. Effect of enriched oxygen atmosphere storage on the quality of live clams (*Ruditapes decussatus*). International Journal of Food Science & Technology 44, 2598–2605. https://doi.org/10.1111/j.1365-2621.2009.02090.x

Gonçalves, A.C., 2010. Qualidade e valorização em aquacultura: propriedades sensoriais e período de conservação útil de peixe e bivalves (PhD Thesis). Universidade de Lisboa, Lisbon, Portugal.

Goulas, A.E., Chouliara, I., Nessi, E., Kontominas, M.G., Savvaidis, I.N., 2005. Microbiological, biochemical and sensory assessment of mussels (*Mytilus galloprovincialis*) stored under modified atmosphere packaging. Journal of Applied Microbiology 98, 752–760. https://doi.org/10.1111/j.1365-2672.2004.02512.x

Goulletquer, P., 2004. Cultured aquatic species information program *Ostrea edulis* [Online]. FAO. Available from: http://www.fao.org/fishery/culturedspecies/Ostrea_edulis/en#tcNA00FE (accessed 10.10.2018).

Green-Petersen, D., Nielsen, J., Hyldig, G., 2012. A model for communication of sensory quality in the seafood processing chain. Critical Reviews in Food Science and Nutrition 52, 443–447. https://doi.org/10.1080/10408398.2010.503907

Grunert, K.G., 2005. Food quality and safety: consumer perception and demand 32. https://doi.org/10.1093/eurrag/jbi011

Guy, A., 2016. The benefits of eating bivalves [Online]. Oceana. Available from: https://oceana.org/blog/benefits-eating-bivalves (accessed 10.11.2018).

Haelters, J., Kerckhof, F., 2009. Background document for *Ostrea edulis* and *Ostrea edulis* beds (Biodiversity Series No. 428/2009). OSPAR.

Harding, J.M., Couturier, C., Parsons, G.J., Ross, N.W., 2004. Evaluation of the neutral red assay as a stress response indicator in cultivated mussels (*Mytilus spp.*) in relation to post-harvest processing activities and storage conditions. Aquaculture 231, 315–326. https://doi.org/10.1016/j.aquaculture.2003.10.025

Hasanspahić, H., 2011. The effects of refrigeration on sensory characteristics of oysters (*Ostrea edulis*). MESO XIII, 369–372.

Hassoun, A., Karoui, R., 2015. Quality evaluation of fish and other seafood by traditional and nondestructive instrumental methods: advantages and limitations. Critical Reviews in Food Science and Nutrition 00–00. https://doi.org/10.1080/10408398.2015.1047926

He, H., 2000. The use of high pressure process to shuck oysters, reduce microbial load and extend shelf-life (Master thesis). Oregon State University, Oregon, EUA.

Hummel, H., de Wolf, L., Zurburg, W., Apon, L., Bogaards, R.H., van Ruitenburg, M., 1989. The glycogen content in stressed marine bivalves: the initial absence of a decrease. Comparative Biochemistry and Physiology Part B: Comparative Biochemistry 94, 729–733. https://doi.org/10.1016/0305-0491(89)90157-0

Hunter, A.C., Weiss, F., Morrison, H., Phillips, J.C., Booth, J.F., Polhamus, L.G., Snapp, O.I., Fortier, S., Harrison, C.W., Alden, C.H., others, 1928. Bacteriology and chemistry of

oysters: with special reference to regulatory control of production, handling, and shipment, A classification of the higher groups and genera of the Coccid Family Margarodidae. U.S. Department of Agriculture.

Idler, D.R., Tamura, T., Wainai, T., 1964. Seasonal variations in the sterol, fat and unsaponifiable components of scallop muscle. Journal of the Fisheries Research Board of Canada 21, 1035–1042. https://doi.org/10.1139/f64-094

IPMA, n.d. Sardinha – *Sardina pilchardus* [Online]. IPMA. Available: http://www.ipma.pt/pt/pescas/recursos/sardinha/?print=true (accessed 9.11.2018).

Jennings, S., Stentiford, G.D., Leocadio, A.M., Jeffery, K.R., Metcalfe, J.D., Katsiadaki, I., Auchterlonie, N.A., Mangi, S.C., Pinnegar, J.K., Ellis, T., Peeler, E.J., Luisetti, T., Baker-Austin, C., Brown, M., Catchpole, T.L., Clyne, F.J., Dye, S.R., Edmonds, N.J., Hyder, K., Lee, J., Lees, D.N., Morgan, O.C., O'Brien, C.M., Oidtmann, B., Posen, P.E., Santos, A.R., Taylor, N.G.H., Turner, A.D., Townhill, B.L., Verner-Jeffreys, D.W., 2016. Aquatic food security: insights into challenges and solutions from an analysis of interactions between fisheries, aquaculture, food safety, human health, fish and human welfare, economy and environment. Fish and Fisheries 17, 893–938. https://doi.org/10.1111/faf.12152

Jiménez-Ruiz, E.I., Márquez-Ríos, E., Cárdenas-López, J.L., Montoya-Camacho, N., Castillo-Yáñez, F.J., Duarte-Figueroa, M.E., Ruiz-Cruz, S., Balois-Morales, R., Ocaño-Higuera, V.M., 2015. Impact of two commercial "in vivo" transport methods on physiological condition of the Japanese oyster (*Crassostrea gigas*). Journal of Chemistry 2015, 1–6. https://doi.org/10.1155/2015/431074

Jiménez-Ruiz, E.I., Ocaño-Higuera, V.M., Maeda-Martínez, A.N., Varela-Romero, A., Márquez-Ríos, E., Muhlia-Almazán, A.T., Castillo-Yáñez, F.J., 2013. Effect of seasonality and storage temperature on rigor mortis in the adductor muscle of lion's paw scallop *Nodipecten subnodosus*. Aquaculture 388–391, 35–41. https://doi.org/10.1016/j.aquaculture.2013.01.006

Karlsen, K.M., n.d. Seafood quality [Online]. NOFIMA. Available from: https://nofima.no/en/prosjekt/seafood-quality/ (accessed 7.11.2018).

Kawashima, K., Yamanaka, H., 1992. Effects of storage temperatures on the post-mortem biochemical changes in scallop adductor muscle. Nippon suisan gakkaishi 58, 2175–2180. https://doi.org/10.2331/suisan.58.2175

Lacoste, A., Jalabert, F., Malham, S.K., Cueff, A., Poulet, S.A., 2001. Stress and stress-induced neuroendocrine changes increase the susceptibility of Juvenile oysters (*Crassostrea gigas*) to *Vibrio splendidus*. Applied and Environmental Microbiology 67, 2304–2309. https://doi.org/10.1128/AEM.67.5.2304-2309.2001

Lemasson, A.J., Kuri, V., Hall-Spencer, J.M., Fletcher, S., Moate, R., Knights, A.M., 2017. Sensory qualities of oysters unaltered by a short exposure to combined elevated pCO2 and temperature. Frontiers in Marine Science 4. https://doi.org/10.3389/fmars.2017.00352

Lerfall, J., Roth, B., Skare, E.F., Henriksen, A., Betten, T., Dziatkowiak-Stefaniak, M.A., Rotabakk, B.T., 2015. Pre-mortem stress and the subsequent effect on flesh quality of pre-rigor filleted Atlantic salmon (*Salmo salar* L.) during ice storage. Food Chemistry 175, 157–165. https://doi.org/10.1016/j.foodchem.2014.11.111

Maguire, J.A., Coleman, A., Jenkins, S., Burnell, G.M., 2002. Effects of dredging on undersized scallops. Fisheries Research 56, 155–165. https://doi.org/10.1016/S0165-7836(01)00322-8

Maguire, J.A., Fleury, P.-G., Burnell, G.M., 1999. Some methods for quantifying quality in the scallop *Pecten maximus* (L.). National Shellfisheries Association 18, 59–66.

Makri, M., 2009. The biochemical textural and sensory properties of frozen stored (-22 C) king scallop *Pecten maximus* meats. African Journal of Biotechnology 8, 3893–3903.

Manalo, A.B., Gempesaw, C.M., 1997. Preferences for oyster attributes by consumers in the U.S. Northeast. Journal of Food Distribution Research 28, 55–63.

Mann, R., 1982. The seasonal cycle of gonadal development in *Arctica islandica* from the southern new england shelf. Fishery bulletin 80.

Marquez Rios, E., Gómez-Jiménez, S., Ocaño-Higuera, V., Castillo-Yáñez, F., Pacheco-Aguilar, R., 2007. Efecto de la emersión a corto plazo a dos temperaturas de exposición aérea sobre parámetros de calidad en cola de langosta espinosa (*Panulirus interruptus*). Ciencias Marinas 33, 73–82.

Martinsdóttir, E., Schelvis, R., Hyldig, G., Sveinsdóttir, K., 2009. Quality safety and authenticity. Sensory evaluation of seafood general principles and guidelines, in: Rehbein, H., Oehlenschläger, J. (Eds.), Fishery Products: Quality, Safety and Authenticity. Wiley-Blackwell Pub., United Kingdom, pp. 411–424.

Matias, M., 2013. Influência da variação sazonal no valor nutricional e avaliação da estabilidade da amêijoa-boa produzida em aquacultura (Master thesis). Instituto Politécnico de Leiria, Leiria, Portugal.

Matthiessen, G.C., 2008. Oyster Culture Fishing News Books Series. John Wiley & Sons, New York, NY.

Maxwell-Miller, G., Josephson, R.V., Spindler, A.A., Holloway-Thomas, D., Avery, M.W., Phleger, C.F., 1982. Chilled (5 ºC) and frozen (-18 ºC) storage stability of the Purple-Hinge Rock scallop, *Hinnites multirugosus* Gale. Journal of Food Science 47, 1654–1661. https://doi.org/10.1111/j.1365-2621.1982.tb05005.x

Mercaldo-Allen, R., Goldberg, R., 2011. Review of the ecological effects of dredging in the cultivation and harvest of molluscan shellfish (Memorandum No. NMFS-NE-220). NOAA, Massachussets, USA.

Merkin, G.V., Roth, B., Gjerstad, C., Dahl-Paulsen, E., Nortvedt, R., 2010. Effect of pre-slaughter procedures on stress responses and some quality parameters in sea-farmed rainbow trout (*Oncorhynchus mykiss*). Aquaculture 309, 231–235. https://doi.org/10.1016/j.aquaculture.2010.08.025

Montanhini, M.T.M., Neto, R.M., 2015. Changes in the microbiological quality of Mangrove oysters (*Crassostrea brasiliana*) during different storage conditions. Journal of Food Protection 78, 164–171. https://doi.org/10.4315/0362-028X.JFP-14-255

Montgomerie, M., 2015. Basic fishing methods.

Mørkøre, T., Mazo T., P.I., Tahirovic, V., Einen, O., 2008. Impact of starvation and handling stress on rigor development and quality of Atlantic salmon, *Salmon salar* L. Aquaculture 277, 231–238. https://doi.org/10.1016/j.aquaculture.2008.02.036

Morton, B., 2018. Bivalve. Encyclopedia Britannica.

Mota, F.C., 2013. Estudo da deterioração da amêijoa-boa *Ruditapes decussatus* (Linnaeus, 1758) (Master thesis). Universidade do Algarve, Algarve, Portugal.

Mueller Loose, S., Peschel, A., Grebitus, C., 2013. Quantifying effects of convenience and product packaging on consumer preferences and market share of seafood products: The case of oysters. Food Quality and Preference 28, 492–504. https://doi.org/10.1016/j.foodqual.2012.11.004

Nielsen, J., Hyldig, G., Larsen, E., 2002. 'Eating quality' of fish - A review. Journal of Aquatic Food Product Technology 11, 125–141. https://doi.org/10.1300/J030v11n03_10

Ojea, J., Pazos, A.J., Martínez, D., Novoa, S., Sánchez, J.L., Abad, M., 2004. Seasonal variation in weight and biochemical composition of the tissues of *Ruditapes decussatus* in relation to the gametogenic cycle. Aquaculture 238, 451–468. https://doi.org/10.1016/j.aquaculture.2004.05.022

Olafsdóttir, 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, 258–265. https://doi.org/10.1016/S0924-2244(97)01049-2

Otoni, C.G., Espitia, P.J.P., Avena-Bustillos, R.J., McHugh, T.H., 2016. Trends in antimicrobial food packaging systems: Emitting sachets and absorbent pads. Food Research International 83, 60–73. https://doi.org/10.1016/j.foodres.2016.02.018

Otwell, S., Garrido, L., Garrido, V., Sims, C., 2011. Assessment studies for Post-Harvest (PHP) oysters. Interstate Shellfish Sanitation Conference (ISSC).

Overaa, T., 2001. Live transport of the Great scallop *Pecten maximus*, in: Paust, B.C., Rice, A. (Eds.), Marketing and Shipping Live Aquatic Products: Proceedings of the Second International Conference and Exhibition, November 1999, Seattle, Washington, Report. University of Alaska Sea Grant College Program, Fairbanks, Alaska, pp. 105–109.

Perry, F., Jackson, A., 2017. *Ostrea edulis* native oyster [Online]. MarLin. Available from: https://www.marlin.ac.uk/species/detail/1146 (accessed 11.13.2017).

Refaey, M.M., Tian, X., Tang, R., Li, D., 2017. Changes in physiological responses, muscular composition and flesh quality of channel catfish *Ictalurus punctatus* suffering from transport stress. Aquaculture 478, 9–15. https://doi.org/10.1016/j.aquaculture.2017.01.026

Roth, B., Birkeland, S., Oyarzun, F., 2009. Stunning, pre slaughter and filleting conditions of Atlantic salmon and subsequent effect on flesh quality on fresh and smoked fillets. Aquaculture 289, 350–356. https://doi.org/10.1016/j.aquaculture.2009.01.013

Ruiz-Capillas, C., Horner, W., Gillyon, C., 2001. Effect of packaging on the spoilage of king scallop *Pecten maximus* during chilled storage. European Food Research and Technology 213, 95–98. https://doi.org/10.1007/s002170100342

Sadok, S., Uglow, R.F., Amor, E.A., 2004. Nitrogenous compound changes in live, stored clam, *Tapes decussatus*. Journal of Aquatic Food Product Technology 12, 113–128. https://doi.org/10.1300/J030v12n04_08

Schreck, C.B., Tort, L., 2016. The concept of stress in fish, in: Fish Physiology. Elsevier, pp. 1–34. https://doi.org/10.1016/B978-0-12-802728-8.00001-1

Seafish, 2013. Responsible sourcing guide: scallops.

Seafish, 1997. Guidelines for the harvesting, handling and distribution of live bivalve molluscs.

Sealife, n.d. *Arctica islandica* Linnaeus, 1767 [Online]. SeaLifeBase. Available from: https://www.sealifebase.ca/summary/Arctica-islandica.html (accessed 11.10.2018)

Seaman, M.N.L., 1991. Survival and aspects of metabolism in oysters, *Crassostrea gigas*, during and after prolonged air storage. Aquaculture 93, 389–395. https://doi.org/10.1016/0044-8486(91)90228-Y

Selye, H., 1973. The evolution of the stress concept. American scientist 61, 692–699. https://doi.org/10.1016/0002-9149(70)90796-4

Serio, A., Fusella, G.C., Chaves López, C., Sacchetti, G., Paparella, A., 2014. A survey on bacteria isolated as hydrogen sulfide-producers from marine fish. Food Control 39, 111–118. https://doi.org/10.1016/j.foodcont.2013.11.003

Silva, H., Costa, P., Rodrigues, S., n.d. Morfologia, biologia e ecologia dos moluscos bivalves. Enciclopedia IPMA.PT.

Steine, G., Alfnes, F., Rørå, M.B., 2005. The effect of color on consumer WTP for farmed salmon. Marine Resource Economics 20, 211–219. https://doi.org/10.1086/mre.20.2.42629470

Stemmer, K., 2013. Shell formation and microstructure of the ocean quahog *Arctica islandica*: Does ocean acidification matter? (PhD Thesis). Universität Bremen, Bremen.

Strohmeier, T., Duinker, A., Lie, Ø., 2000. Seasonal variations in chemical composition of the female gonad and storage organs in *Pecten maximus* (L.) suggesting that somatic and reproductive growth are separated in time. Journal of Shellfish Research 19, 741–747.

Svåsand, T., Crosetti, D., García-Vázquez, E., Verspoor, E., 2007. Genetic impact of aquaculture activities on native populations (Genimpact final scientific report No. EU contract n. RICA-CT-2005-022802).

Thorarinsdóttir, G., Steingrímsson, S.A., 2000. Size and age at sexual maturity and sex ratio in ocean quahog *Arctica islandica* (Linneaus, 1767), of north-west Iceland. Journal of Shellfish Research 19, 943–947.

Torgersen, J.S., Koppang, E.O., Stien, L.H., Kohler, A., Pedersen, M.E., Mørkøre, T., 2014. Soft texture of Atlantic salmon fillets is associated with glycogen accumulation. PLoS ONE 9, e85551. https://doi.org/10.1371/journal.pone.0085551

Torres, J., 2011. Efeito de diferentes temperaturas de armazenamento na qualidade de moluscos bivalves vivos (Master thesis). Universidade de Lisboa, Lisbon, Portugal.

Vanhonacker, F., Pieniak, Z., Verbeke, W., 2013. European consumer perceptions and barriers for fresh, frozen, preserved and ready-meal fish products. British Food Journal 115, 508–525. https://doi.org/10.1108/00070701311317810

Venugopal, V., Gopakumar, K., 2017. Shellfish: nutritive value, health benefits, and consumer safety. Comprehensive Reviews in Food Science and Food Safety 16, 1219–1242. https://doi.org/10.1111/1541-4337.12312

Waagbø, R., 2016. Pigments, Presented at the course Fish nutrition, Bergen, 30 September

Wang, J., 2015. Quality evaluation of live Eastern oyster (*Crassostrea virginica*) based on textural profiling analysis, free amino acids analysis, and consumer sensory evaluation. Faculty of Auburn University, Alabama, US.

Widdows, J., Bayne, B.., Livingstone, D.., Newell, R.I.., Donkin, P., 1979. Physiological and biochemical responses of bivalve molluscs to exposure to air. Comparative Biochemistry and Physiology Part A: Physiology 62, 301–308. https://doi.org/10.1016/0300-9629(79)90060-4

Woll, A.K., Bakke, S., 2017. Stress and mortality in the supply chain of live scallops *Pecten maximus* L., from scuba diver to market. Aquaculture Research 48, 594–607. https://doi.org/10.1111/are.12906

Zhang, J., Walton, W.C., Wang, Y., 2017. Quantitative quality evaluation of Eastern oyster (*Crassostrea virginica*) cultured by two different methods. Aquaculture Research 48, 2934–2944. https://doi.org/10.1111/are.13126

7. Appendices

Appendix 1: Surveys

Figure 1a: Survey for the sensory evaluation of Oysters. The survey has two types of parameters: one for the smell characteristics and another for appearance characteristics. Each characteristic have to be evaluated by a level of intensity, ranged from level 0 to level 5. Also, in the end there is a question which should be answer after evaluate all the parameters and consider all the characteristics of the sample.

Assessor: _____ **Oyster code:** _____

Parameters			**Intensity**					
			0	1	2	3	4	5
			Absent	Very weak	Weak	Medium	Intense	Very intense
Smell	Fresh, algal, seawater							
	Sour							
	Ammonia							
	Sulfhydric, putrid							
Appearance	Bright surface							
	Body muscle dehydrated							
	Plum *Plomme*	White						
		Well rounded						
	Gills	Intact						
	Intervalval fluid	Transparent						
		Sufficient quantity						

1) Would you eat this animal?

Yes ____ No ____

Figure 1b: Survey for the sensory evaluation of scallops. The survey has two types of parameters: one for the smell characteristics and another for appearance characteristics. Each characteristic have to be evaluated by a level of intensity, ranged from level 0 to level 5. Also, in the end there is a question which should be answer after evaluate all the parameters and consider all the characteristics of the sample.

Assessor: _____ **Scallop code**: _____

Parameters		**Intensity**					
		0	1	2	3	4	5
		Absent	Very weak	Weak	Medium	Intense	Very intense
Smell	Sulfhydric, putrid						
	Ammonia						
	Sour						
	Musty						
	Boiled milk, seaweed						
	Fresh, seawater						
Appearance	Slight blackening						
	Brownish color						
	Slimy						
	Bright surface						

1) Would you eat this animal?

Yes ____ No ____

Figure 1c: Survey for the sensory evaluation of Clams. The survey has two types of parameters: one for the smell characteristics and another for appearance characteristics. Each characteristic have to be evaluated by a level of intensity, ranged from level 0 to level 5. Also, in the end there is a question which should be answer after evaluate all the parameters and consider all the characteristics of the sample.

Assessor: _____ **Clam code:** _____

Parameters		**Intensity**					
		0	1	2	3	4	5
		Absent	Very weak	Weak	Medium	Intense	Very intense
Smell	Fresh, algal, seawater						
	Sour						
	Ammonia						
	Sulfhydric, putrid						
Appearance	Bright surface						
	Cream color						
	White color						
	Brownish color						

Would you eat this animal?

Yes ____ No ____

Appendix 2: Data

Table 2a: The average of the weight losses of small and big scallops during the storage at different temperatures (3, 6 and 9 ºC) until 12 days.

Temperature ºC	Day	Small		Big	
		Average weight lost (gr.)	% weight lost	Average weight lost (gr.)	% weight lost
3	3	15.1	6.9	65.7	19.9
3	6	20.0	9.2	56.2	17.0
3	9	23.9	11.0	66.5	20.1
3	12	21.1	9.7	54.2	16.4
6	3	23.6	10.8	33.5	10.7
6	6	31.7	14.5	50.8	16.2
6	9	32.5	14.8	68.3	21.8
6	12	30.2	13.8	76.6	24.4
9	3	23.4	11.0	40.5	12.3
9	6	27.2	12.8	53.3	16.2
9	9	28.8	13.6	77.2	23.5
9	12	15.5	7.3	91.6	27.9

Table 2b: The average of the weight losses (in grams and %) of small and big oysters during the storage at different temperatures (3, 6 and 9 ºC) until 12 days.

Temperature ºC	Day	Small		Big	
		Average weight lost (gr.)	% weight lost	Average weight lost (gr.)	% weight lost
3	3	0.6	1.1	5.5	3.1
3	6	0.9	1.7	5.1	2.9
3	10	0.0	0.0	9.1	5.1
3	12	2.0	3.6	17.4	9.9
6	3	1.6	2.9	9.4	5.3
6	6	2.3	4.1	12.4	6.9
6	10	3.3	5.8	21.1	11.9
6	12	3.6	6.3	34.8	19.5
9	3	2.1	3.7	6.9	3.8
9	6	3.7	6.6	14.6	8.1
9	10	4.2	7.4	17.0	9.4
9	12	3.9	6.8	21.0	11.7

Table 2c: The average of the weight losses of clams during the storage at different temperatures (3, 6 and 9 ºC) until 12 days.

Temperature ºC	Day	Average weight lost (gr)	% weight lost
3	3	5.5	2.1
3	6	4.5	1.7
3	9	27.4	10.4
3	12	71.2	27.6
6	3	5.4	2.0
6	6	5.2	1.9
6	9	43.3	16.2
6	12	61.5	22.5
9	3	16.1	6.3
9	6	11.8	4.6
9	9	15.0	6.3
9	12	48.2	21.2

Table 2d: Number of scallops which survived during the storage at different temperatures (3, 6 and 9 ºC). Each sample had a total of 7 scallops.

	Scallops small			Scallops big			
Temperature / Day	3ºC	6ºC	9ºC	3ºC	6ºC	9ºC	Total (survivors/total scallops)
3 days	6	4	5	7	7	4	33
7 days	6	0	0	6	0	0	12
10 days	3	0	0	0	0	0	3
12 days	0	0	0	0	0	0	0
Total	15	4	5	13	7	4	168

Table 2e: Acceptance of oysters by the participants. The number of participants who accepted the small oysters and big oysters is in number and in percentage (%).

Season	Temperature ºC	Size	Days	Nº of participants who accepted the sample	% of participants who accepted the sample
March	3	small	3	4	100
March	3	small	6	4	100
March	3	small	9	4	100
March	3	small	12	3	75
March	3	big	6	4	100
March	3	big	9	3	75
March	3	big	12	1	25
June	3	small	3	N/A	NA
June	3	small	6	2	66.7
June	3	small	9	0	0
June	3	big	3	N/A	NA
June	3	big	6	2	66.7
June	3	big	9	1	33.3
August	3	small	3	4	66.7
August	3	small	6	2	33.3
August	3	small	9	2	33.3
August	3	big	3	4	66.7
August	3	big	6	5	83.3
August	3	big	9	4	66.7
March	6	small	3	2	50
March	6	small	6	4	100
March	6	small	9	4	100
March	6	small	12	4	100
March	6	big	6	0	0
March	6	big	9	2	50
March	6	big	12	4	100
June	6	small	3	N/A	NA
June	6	small	6	2	66.7
June	6	small	9	1	33.3
June	6	big	3	N/A	NA
June	6	big	6	3	100
June	6	big	9	0	0
August	6	small	3	4	66.7
August	6	small	6	3	50
August	6	small	9	0	0
August	6	big	3	5	83.3
August	6	big	6	5	83.3

August	6	big	9	5	83.3
March	9	small	3	1	25
March	9	small	6	4	100
March	9	small	9	4	100
March	9	small	12	0	0
March	9	big	6	0	0
March	9	big	9	4	100
March	9	big	12	4	100
June	9	small	3	1	33.3
June	9	small	6	2	66.7
June	9	small	9	0	0.0
June	9	big	3	1	33.3
June	9	big	6	3	100
June	9	big	9	3	100
August	9	small	3	1	16.7
August	9	small	6	4	66.7
August	9	small	9	4	66.7
August	9	big	3	4	66.7
August	9	big	6	5	83.3
August	9	big	9	4	66.7

Table 2f: Acceptance of scallops by the participants. The number of participants who accepted the small scallops and big scallops is in number and in percentage (%).

Season	Temperature ºC	Type	Days	Nº of participants who accepted the sample	% of participants who accepted the sample
March	3	small	3	4	100
March	3	big	3	4	100
March	3	small	6	0	0
March	3	big	6	4	100
March	3	small	9	0	0
March	3	big	9	1	25
March	6	small	3	4	100
March	6	big	3	4	100
March	6	small	6	0	0
March	6	big	6	0	0
March	6	small	9	0	0
March	6	big	9	0	0
March	9	small	3	3	75
March	9	big	3	3	75
March	9	small	6	0	0
March	9	big	6	0	0
March	9	small	9	0	0
March	9	big	9	0	0
June	3	big	3	3	100
June	3	cover	3	2	66.7
June	3	diaper	3	2	66.7
June	3	small	6	2	66.7
June	3	big	6	1	33.3
June	3	cover	6	3	100
June	3	diaper	6	2	66.7
June	3	small	9	0	0
June	3	big	9	0	0
June	3	cover	9	0	0
June	3	diaper	9	0	0
June	6	big	3	3	100
June	6	cover	3	3	100
June	6	diaper	3	3	100
June	6	small	6	0	0
June	6	big	6	1	33.3
June	6	cover	6	1	33.3
June	6	diaper	6	0	0
June	6	small	9	0	0

June	6	big	9	0	0
June	6	cover	9	0	0
June	6	diaper	9	0	0
June	9	small	3	2	66.7
June	9	big	3	3	100
June	9	cover	3	3	100
June	9	diaper	3	3	100
June	9	cover	6	0	0
June	9	diaper	6	0	0
June	9	cover	9	0	0
June	9	diaper	9	0	0
August	3	small	3	6	100
August	3	big	3	6	100
August	3	small	6	4	66.7
August	3	big	6	4	66.7
August	6	small	3	5	83.3
August	6	big	3	5	83.3
August	6	small	6	0	0
August	6	big	6	2	33.3

Table 2g: Acceptance of clams by the participants. The number of participants who accepted the clams is in number and in percentage (%).

Season	Temperature ºC	Days	Nº of participants who accepted the sample	% of participants who accepted the sample
March	3	9	4	100
March	3	12	3	75
March	6	9	3	75
March	6	12	4	100
March	9	9	4	100
March	9	12	2	50
June	3	9	3	100
June	6	9	3	100
June	9	9	3	100

Appendix 3: Statistical analyzes

3.1 Oysters

Table 3a: Statistical data of oysters with the *p-value* per parameter and *p-value* for all the predictors variables.

Parameter		***p-value* (parameter)**	***p-value* (predictor variable)**			
			Season	**Temperature**	**Size**	**Day**
Fresh, algal, seawater		0.05708	0.04692	0.03403	0.9195	0.5836
Sour		3.972e-05	9.603e-06	0.481	0.8526	0.0002167
Ammonia		0.1056	0.06161	0.1709	0.572	0.07102
Sulfhydric, putrid		0.0004275	1.177e-05	0.6415	0.6056	0.0143
Bright surface		0.01196	0.06562	0.2772	0.006369	0.3894
Body muscle dehydrated		0.9192	0.6126	0.934	0.3229	0.6576
Plum	White creamy	1.988e-06	5.442e-07	0.6948	0.02668	0.006789
	Well rounded	1.074e-08	2.764e-07	0.9459	0.0003531	0.001536
Gills	Intact	0.01245	0.001822	0.2946	0.4385	0.09046
Intervalval fluid	Transparent	0.001171	0.0008094	0.2264	0.156	0.812
	Sufficient quantity	0.01202	0.003486	0.3624	0.09849	0.6956

Statistical data: Quality overall vision

ANOVA

```
            Df  Sum Sq Mean Sq F value  Pr(>F)
Size         1   2.294 2.29416  4.1436 0.04295 *
Day          1   0.011 0.01097  0.0198 0.88816
Temp         1   0.596 0.59642  1.0772 0.30042
Season       2   2.720 1.36007  2.4565 0.08801 .
Residuals  227 125.681 0.55366
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1
```

Summary

```
Residuals:
     Min       1Q   Median       3Q      Max
-2.01705 -0.50341  0.05825  0.53286  1.57055

Coefficients:
             Estimate Std. Error t value Pr(>|t|)
(Intercept)   3.73194    0.23365  15.972   <2e-16 ***
Sizesmall    -0.21950    0.09844  -2.230   0.0267 *
Day          -0.01244    0.01882  -0.661   0.5091
Temp         -0.01925    0.01987  -0.969   0.3336
SeasonJune   -0.30694    0.14541  -2.111   0.0359 *
SeasonAugust -0.19391    0.12025  -1.613   0.1082
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 0.7441 on 227 degrees of freedom
  (1 observation deleted due to missingness)
Multiple R-squared:  0.04281,  Adjusted R-squared:  0.02173
F-statistic: 2.031 on 5 and 227 DF,  p-value: 0.07528
```

Multiple comparisons: Tukey HSD

Quality x Season

ANOVA

```
             Df Sum Sq Mean Sq F value Pr(>F)
Season        2   2.29  1.1452   2.042  0.132
Residuals   230 129.01  0.5609
1 observation deleted due to missingness
```

TukeyHSD

```
  Tukey multiple comparisons of means
    95% family-wise confidence level

Fit: aov(formula = mean_score ~ Season, data = oyster.df2)

$Season
                   diff        lwr       upr     p adj
June-March   -0.2764069 -0.6102929 0.0574790 0.1264443
August-March -0.1432505 -0.4008009 0.1142999 0.3898895
August-June   0.1331564 -0.1885455 0.4548584 0.5925113
```

Quality x Size

ANOVA

```
             Df Sum Sq Mean Sq F value Pr(>F)
Size          1   2.29  2.2942   4.108 0.0438 *
Residuals   231 129.01  0.5585
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1
1 observation deleted due to missingness
```

TukeyHSD

```
  Tukey multiple comparisons of means
    95% family-wise confidence level

Fit: aov(formula = mean_score ~ Size, data = oyster.df2)

$Size
                diff        lwr          upr     p adj
small-big -0.1986775 -0.3918161 -0.005538965 0.0438326
```

Statistical data: acceptance of oysters by the participants

ANOVA

```
             Df Deviance Resid. Df Resid. Dev  Pr(>Chi)
NULL                           226     295.81
Season        2   7.5603       224     288.25 0.0228195 *
Size          1   3.2815       223     284.97 0.0700646 .
Season:Size   2  14.6954       221     270.28 0.0006441 ***
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1
```

Summary

```
Deviance Residuals:
    Min       1Q   Median       3Q      Max
-1.9291  -1.1127   0.5815   0.9282   1.3893

Coefficients:
                       Estimate Std. Error z value Pr(>|z|)
(Intercept)             0.52609    0.34983   1.504 0.132614
SeasonJune              0.09295    0.58494   0.159 0.873749
SeasonAugust            0.62253    0.47296   1.316 0.188093
Sizesmall               1.16558    0.53995   2.159 0.030876 *
SeasonJune:Sizesmall   -2.27013    0.84454  -2.688 0.007188 **
SeasonAugust:Sizesmall -2.46836    0.68574  -3.600 0.000319 ***
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

(Dispersion parameter for binomial family taken to be 1)

    Null deviance: 295.81  on 226  degrees of freedom
Residual deviance: 270.28  on 221  degrees of freedom
  (7 observations deleted due to missingness)
AIC: 282.28

Number of Fisher Scoring iterations: 4
```

Statistical data: Weight lost

ANOVA

```
              Df  Sum Sq Mean Sq F value    Pr(>F)
size           1  75.397  75.397 15.2860 0.0007514 ***
day            1 298.293 298.293 60.4761 9.415e-08 ***
temp           1  45.070  45.070  9.1375 0.0062544 **
size:day       1  52.976  52.976 10.7404 0.0034427 **
size:temp      1   1.812   1.812  0.3673 0.5506975
day:temp       1   9.587   9.587  1.9436 0.1771962
size:day:temp  1   0.516   0.516  0.1046 0.7494443
Residuals     22 108.513   4.932
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1
```

Summary

```
Residuals:
    Min      1Q  Median      3Q     Max
-3.5786 -0.7434  0.2269  0.7578  7.1025

Coefficients:
                  Estimate Std. Error t value Pr(>|t|)
(Intercept)       -0.70549    2.62147  -0.269   0.7903
sizeSmall          0.27573    3.70732   0.074   0.9414
day                0.77697    0.34481   2.253   0.0345 *
temp               0.15031    0.40450   0.372   0.7137
sizeSmall:day     -0.75003    0.48764  -1.538   0.1383
sizeSmall:temp     0.04977    0.57205   0.087   0.9315
day:temp           0.04028    0.05321   0.757   0.4570
sizeSmall:day:temp 0.02433    0.07524   0.323   0.7494
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 2.221 on 22 degrees of freedom
Multiple R-squared:  0.8168,   Adjusted R-squared:  0.7584
F-statistic: 14.01 on 7 and 22 DF,  p-value: 8.605e-07
```

Statistical data: Survival

ANOVA

```
              Df Sum Sq Mean Sq F value  Pr(>F)
size           1 136.14 136.136  4.1558 0.05837 .
days           1 108.91 108.909  3.3247 0.08698 .
temp           1  51.05  51.051  1.5584 0.22985
size:days      1   6.81   6.807  0.2078 0.65463
size:temp      1   0.00   0.000  0.0000 1.00000
days:temp      1  91.89  91.892  2.8052 0.11339
size:days:temp 1   0.00   0.000  0.0000 1.00000
Residuals     16 524.12  32.758
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1
```

Summary

```
Residuals:
     Min       1Q   Median       3Q      Max
-11.9083  -1.8458   0.4763   3.8702   7.1450

Coefficients:
                     Estimate Std. Error t value Pr(>|t|)
(Intercept)          8.095e+01  1.071e+01   7.560 1.15e-06 ***
sizesmall           -7.145e+00  1.514e+01  -0.472    0.643
days                 1.905e+00  1.303e+00   1.462    0.163
temp                 2.382e+00  1.652e+00   1.441    0.169
sizesmall:days       3.176e-01  1.843e+00   0.172    0.865
sizesmall:temp      -2.767e-14  2.337e+00   0.000    1.000
days:temp           -2.382e-01  2.011e-01  -1.184    0.254
sizesmall:days:temp  2.736e-15  2.844e-01   0.000    1.000
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 5.723 on 16 degrees of freedom
Multiple R-squared:  0.4296,   Adjusted R-squared:  0.1801
F-statistic: 1.722 on 7 and 16 DF,  p-value: 0.1741
```

3.2 Scallops

Table 3b: Statistical data of scallops with the *p-value* per parameter and *p-value* for all the predictors variables.

Parameter	*p-value* (parameter)	*p-value* (predictor variable)			
		Season	Temperature	Type	Day
Sulfhydric, putrid	2.2e-16	0.000242	1.372e-05	0.3619	2.2e-16
Ammonia	2.2e-16	4.63e-05	2.053e-05	0.4133	2.2e-16
Sour	2.2e-16	2.146e-05	0.0004155	0.5071	2.2e-16
Musty	2.2e-16	1.039e-10	0.01676	0.0007559	3.655e-07
Boiled milk, seaweed	8.966e-05	0.05123	0.1108	0.9867	9.132e-07
Fresh, seawater	2.2e-16	6.34e-06	0.0155	0.6637	2.2e-16
Slight blackening	3.691e-09	0.0071	0.009175	0.2728	1.119e-07
Brownish color	1.691e-14	0.7769	0.008621	0.9565	3.563e-15
Slimy	1.093e-12	0.1243	0.09586	0.9309	7.022e-14
Bright surface	3.145e-09	0.04623	0.01185	0.5669	4.185e-08

Statistical data: Quality overall vision

ANOVA

```
Response: mean_score
           Df  Sum Sq Mean Sq  F value    Pr(>F)
Temp        1  38.025  38.025  74.6406 1.840e-15 ***
Type        3   4.906   1.635   3.2099   0.02411 *
Season      2  18.205   9.103  17.8679 7.298e-08 ***
Day         1 177.365 177.365 348.1575 < 2.2e-16 ***
Residuals 199 101.379   0.509
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1
```

Summary

```
Call:
lm(formula = mean_score ~ Temp + Type + Season + Day, data = scallop.df2)

Residuals:
    Min      1Q  Median      3Q     Max
-1.7464 -0.3905  0.0672  0.4650  1.9258

Coefficients:
             Estimate Std. Error t value Pr(>|t|)
(Intercept)   5.97658    0.21865  27.334  < 2e-16 ***
Temp         -0.19547    0.02248  -8.694  1.3e-15 ***
Typecover    -0.03248    0.18882  -0.172 0.863594
Typediaper   -0.04730    0.18882  -0.250 0.802473
Typesmall    -0.20511    0.11583  -1.771 0.078121 .
SeasonJune    0.56223    0.14775   3.805 0.000188 ***
SeasonAugust  0.14145    0.14214   0.995 0.320865
Day          -0.41182    0.02207 -18.659  < 2e-16 ***
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 0.7138 on 199 degrees of freedom
  (3 observations deleted due to missingness)
Multiple R-squared:  0.7017,   Adjusted R-squared:  0.6912
F-statistic: 66.88 on 7 and 199 DF,  p-value: < 2.2e-16
```

Multiple comparisons: Tukey HSD

Quality x Season

ANOVA

```
             Df Sum Sq Mean Sq F value  Pr(>F)
Season        2  31.51  15.755   10.42 4.9e-05 ***
Residuals   204 308.37   1.512
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1
3 observations deleted due to missingness
```

TukeyHSD

```
  Tukey multiple comparisons of means
    95% family-wise confidence level

Fit: aov(formula = mean_score ~ Season, data = scallop.df2)

$Season
                  diff        lwr      upr     p adj
June-March   0.6289234  0.1644503 1.093397 0.0045713
August-March 1.0160067  0.4704303 1.561583 0.0000524
August-June  0.3870833 -0.1317233 0.905890 0.1852562
```

Quality x Type

ANOVA

```
             Df Sum Sq Mean Sq F value Pr(>F)
Type          3    3.3   1.109   0.669  0.572
Residuals   203  336.6   1.658
3 observations deleted due to missingness
```

TukeyHSD

```
  Tukey multiple comparisons of means
    95% family-wise confidence level
                    diff        lwr       upr     p adj
cover-big    -0.01575213 -0.7581156 0.7266113 0.9999402
diaper-big   -0.03056695 -0.7729304 0.7117965 0.9995644
small-big    -0.27388957 -0.8137617 0.2659825 0.5549648
diaper-cover -0.01481481 -0.9226033 0.8929737 0.9999728
small-cover  -0.25813744 -1.0094286 0.4931537 0.8100343
small-diaper -0.24332262 -0.9946138 0.5079685 0.8358154
```

Statistical data: acceptance of scallops by the participants

ANOVA

```
     Df Deviance Resid. Df Resid. Dev Pr(>Chi)
NULL                   199     276.94
Day   1  146.855       198     130.08   <2e-16 ***
Type  3    4.952       195     125.13   0.1754
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1
```

Summary

```
Deviance Residuals:
    Min       1Q   Median       3Q      Max
-2.6758  -0.6735  -0.1030   0.3162   2.8990

Coefficients:
            Estimate Std. Error z value Pr(>|z|)
(Intercept)   7.4210     1.1231   6.608 3.90e-11 ***
Day          -1.2898     0.1849  -6.975 3.07e-12 ***
Typecover     0.1227     0.7147   0.172   0.8637
Typediaper   -0.5812     0.7735  -0.751   0.4524
Typesmall    -1.0504     0.5304  -1.980   0.0477 *
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

(Dispersion parameter for binomial family taken to be 1)

    Null deviance: 276.94  on 199  degrees of freedom
Residual deviance: 125.13  on 195  degrees of freedom
  (10 observations deleted due to missingness)
AIC: 135.13

Number of Fisher Scoring iterations: 6
```

Statistical data: Weight lost

ANOVA

```
              Df Sum Sq Mean Sq F value    Pr(>F)
size           1 215.35  215.35 13.0911  0.001523 **
day            1 904.77  904.77 55.0008 2.028e-07 ***
temp           1  14.37   14.37  0.8734  0.360165
size:day       1 122.98  122.98  7.4760  0.012110 *
size:temp      1   0.09    0.09  0.0052  0.943097
day:temp       1   4.29    4.29  0.2607  0.614714
size:day:temp  1  15.21   15.21  0.9243  0.346791
Residuals     22 361.90   16.45
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1
```

Summary

```
Residuals:
    Min      1Q  Median      3Q     Max
-6.5365 -2.8053  0.6976  2.3159  7.6588

Coefficients:
                   Estimate Std. Error t value Pr(>|t|)
(Intercept)         5.72032    4.78740   1.195   0.2449
sizesmall          -4.30516    6.77040  -0.636   0.5314
day                 1.10145    0.62970   1.749   0.0942 .
temp               -0.32270    0.73871  -0.437   0.6665
sizesmall:day      -0.12765    0.89053  -0.143   0.8873
sizesmall:temp      0.77543    1.04470   0.742   0.4658
day:temp            0.10114    0.09717   1.041   0.3092
sizesmall:day:temp -0.13211    0.13741  -0.961   0.3468
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 4.056 on 22 degrees of freedom
Multiple R-squared:  0.7792,   Adjusted R-squared:  0.7089
F-statistic: 11.09 on 7 and 22 DF,  p-value: 6.023e-06
```

Statistical data: Survival

ANOVA

```
               Df  Sum Sq Mean Sq F value    Pr(>F)
size            1     0.0     0.0  0.0000   1.00000
days            1 19825.0 19825.0 32.0910 3.516e-05 ***
temp            1  4607.0  4607.0  7.4574   0.01481 *
size:days       1   245.4   245.4  0.3972   0.53743
size:temp       1    12.8    12.8  0.0207   0.88743
days:temp       1  1125.8  1125.8  1.8223   0.19583
size:days:temp  1   207.0   207.0  0.3351   0.57070
Residuals      16  9884.4   617.8
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1
```

Summary

```
Residuals:
    Min      1Q  Median      3Q     Max
-42.850 -14.998  -1.425  15.001  32.853

Coefficients:
                    Estimate Std. Error t value Pr(>|t|)
(Intercept)         185.7333    46.4996   3.994  0.00104 **
sizesmall           -42.9000    65.7603  -0.652  0.52343
days                -16.6689     5.6597  -2.945  0.00951 **
temp                -14.2917     7.1750  -1.992  0.06374 .
sizesmall:days        6.1967     8.0041   0.774  0.45011
sizesmall:temp        4.7667    10.1470   0.470  0.64487
days:temp             1.1911     0.8733   1.364  0.19148
sizesmall:days:temp  -0.7150     1.2351  -0.579  0.57070
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 24.86 on 16 degrees of freedom
Multiple R-squared:  0.7247,   Adjusted R-squared:  0.6043
F-statistic: 6.018 on 7 and 16 DF,  p-value: 0.001455
```

3.3 Clams

Table 3c: Statistical data of clams with the *p-value* per parameter and p-value for all the predictors variables.

Parameter	***p-value* (parameter)**	***p-value* (predictor variable)**		
		Season	**Temperature**	**Day**
Fresh, algal, seawater	0.4445	0.5673	0.4989	0.3227
Sour	0.1571	0.1207	0.6497	0.01838
Ammonia	0.3463	0.3974	0.5892	0.06149
Sulfhydric, putrid	0.2657	0.001772	0.8473	0.02397
Bright surface	0.7	0.5268	0.4552	0.4506
Cream color	0.002517	0.1492	0.4124	0.5308
White color	0.1588	0.382	0.121	0.1343
Brownish color	4.232e-05	4.281e-07	0.8367	0.0178

Statistical data: Quality overall vision

ANOVA

```
            Df Sum Sq Mean Sq F value    Pr(>F)
day          1 2.0050 2.00496 14.2134 0.0008105 ***
temp         1 0.6679 0.66793  4.7350 0.0384768 *
season       1 0.3581 0.35813  2.5389 0.1227165
day:temp     1 0.1050 0.10498  0.7442 0.3959101
temp:season  1 0.0450 0.04501  0.3191 0.5768040
Residuals   27 3.8087 0.14106
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1
```

Summary

```
Residuals:
     Min       1Q   Median       3Q      Max
-0.88889 -0.23958 -0.00521  0.26042  0.60069

Coefficients: (2 not defined because of singularities)
                   Estimate Std. Error t value Pr(>|t|)
(Intercept)         5.84722    1.43428   4.077 0.000361 ***
day                -0.26157    0.13522  -1.934 0.063612 .
temp               -0.16840    0.22131  -0.761 0.453298
seasonJune          0.03472    0.43818   0.079 0.937424
day:temp            0.02141    0.02087   1.026 0.313910
day:seasonJune           NA         NA      NA       NA
temp:seasonJune     0.03819    0.06761   0.565 0.576804
day:temp:seasonJune      NA         NA      NA       NA
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 0.3756 on 27 degrees of freedom
Multiple R-squared:  0.4551,   Adjusted R-squared:  0.3542
F-statistic:  4.51 on 5 and 27 DF,  p-value: 0.004067
```

Statistical data: acceptance of clams by the participants

ANOVA

```
     Df Deviance Resid. Df Resid. Dev Pr(>Chi)
NULL                    30     14.832
day   1   4.4004        29     10.431  0.03593 *
temp  1   0.0321        28     10.399  0.85789
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1
```

Summary

```
Deviance Residuals:
     Min        1Q    Median        3Q       Max
-1.91984   0.00004   0.00005   0.58720   0.68873

Coefficients:
             Estimate Std. Error z value Pr(>|z|)
(Intercept)  7.817e+01  1.584e+04   0.005    0.996
day         -6.360e+00  1.320e+03  -0.005    0.996
temp        -5.874e-02  3.281e-01  -0.179    0.858

(Dispersion parameter for binomial family taken to be 1)

    Null deviance: 14.831  on 30  degrees of freedom
Residual deviance: 10.399  on 28  degrees of freedom
  (2 observations deleted due to missingness)
AIC: 16.399

Number of Fisher Scoring iterations: 19
```

Statistical data: Weight lost

ANOVA

```
          Df Sum Sq Mean Sq F value  Pr(>F)
day        1 909.89  909.89 37.4784 7.5e-05 ***
temp       1   1.16    1.16  0.0477  0.8312
day:temp   1  22.61   22.61  0.9315  0.3552
Residuals 11 267.06   24.28
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1
```

Summary

```
Residuals:
   Min     1Q Median     3Q    Max
-6.862 -3.939  1.044  3.111  5.954

Coefficients:
            Estimate Std. Error t value Pr(>|t|)
(Intercept)  -6.4035     5.8300  -1.098  0.29550
day           2.5446     0.7934   3.207  0.00834 **
temp          0.5955     0.8996   0.662  0.52162
day:temp     -0.1181     0.1224  -0.965  0.35522
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 4.927 on 11 degrees of freedom
Multiple R-squared:  0.7776,   Adjusted R-squared:  0.7169
F-statistic: 12.82 on 3 and 11 DF, p-value: 0.0006525
```

Statistical data: Survival

ANOVA

```
          Df Sum Sq Mean Sq F value Pr(>F)
days       1 115.79 115.787  1.4184 0.2678
temp       1  34.74  34.736  0.4255 0.5325
days:temp  1   6.95   6.947  0.0851 0.7779
Residuals  8 653.04  81.630
```

Summary

```
Residuals:
    Min      1Q  Median      3Q     Max
-12.780  -6.598   2.362   5.626  11.113

Coefficients:
             Estimate Std. Error t value Pr(>|t|)
(Intercept)  1.000e+02  1.690e+01   5.916 0.000355 ***
days        -3.704e-01  2.057e+00  -0.180 0.861583
temp        -1.664e-14  2.608e+00   0.000 1.000000
days:temp   -9.261e-02  3.175e-01  -0.292 0.777915
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 9.035 on 8 degrees of freedom
Multiple R-squared:  0.1943,   Adjusted R-squared:  -0.1079
F-statistic: 0.643 on 3 and 8 DF,  p-value: 0.6086
```