



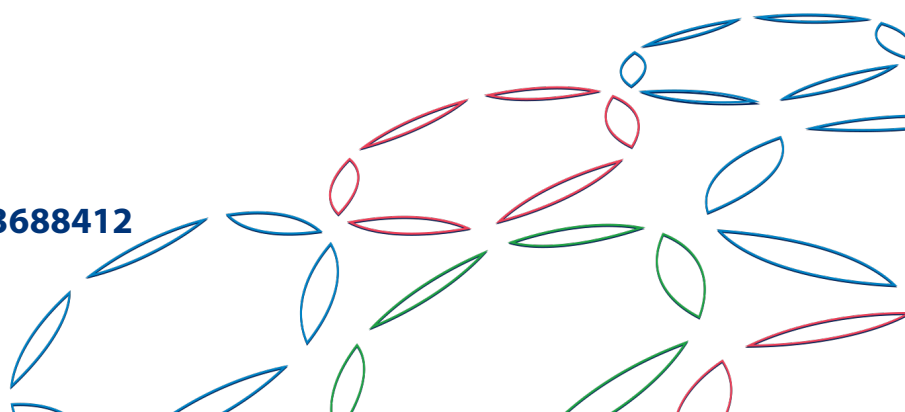
Hámörkun gæða frosinna karfaafurða / Quality optimization of frozen redfish products

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<i>Ágríp á íslensku:</i>	<p>Markmið verkefnisins var tvíþætt. Í fyrsta lagi að rannsaka áhrif tíma og hitastigs við geymslu í frosti, á niðurbrot fitu í karfa. Það var gert með því að bera saman áhrif hitastigsbreytinga og meðhöndlunar í frostgeymslu við flutninga og áhrif á eðlis- og efnaeiginleika ásamt stöðugleika fitu í karfa. Í öðru lagi, að rannsaka áhrif á milli 4ja daga og 9 daga gamals fisks frá veiðum, og mun á milli árstíða á gæði stöðugleika í geymslu á karfa. Geymsluhitastig og tími hafði áhrif á eðlis- og efnaeiginleika karfa, þ.e. á fríar fitusýrur, TBARS og TVB-N. Árstíðamunur hafði áhrif á næringargildi og stöðugleika karfa. Ljósi vöðvinn í karfa sem var veiddur í nóvember innihélt hærra magn af EPA og DHA, en karfi veiddur í júní. Karfi veiddur í nóvember var ekki eins stöðugur í frostgeymslu, þar sem hann innihélt hærra hlutfall af ómettuðum fitusýrum. Ljósi vöðvinn innihélt hærra næringargildi en dökki vöðvinn, sem leiðir til betri uppsprettu næringar fyrir neyslu almennings. Dökki vöðvinn var hins vegar viðkvæmur fyrir oxun fitu, sem gæti haft neikvæð áhrif fyrir ljósa vöðvann. Þá er jafnvel þörf að aðskilja dökka og ljósa vöðvann.</p>		
<i>Lykilorð á íslensku:</i>	<i>Karfi; frostgeymsla; hitastigssveiflur; gæðarýrnun</i>		

<p><i>Summary in English:</i></p>	<p>The aim of the study was twofold. Firstly, to explore the influence of time and temperature during frozen storage on lipid deterioration of red fish. That was done by comparing the effect of temperature fluctuation and abuse during frozen storage, as can be expected during transportation, on the physicochemical characteristics and lipid stability of redfish fillets. Secondly, to investigate the effect of 4 days postcatch and 9 days postcatch, and seasonal variation on the quality and storage stability of frozen red fish.</p> <p>Storage temperature and storage time affected the physical- and chemical properties in redfish, e.g free fatty acids, TBARS and TVB-N. Season of capture affected both the nutritional value and stability of golden redfish. The light muscle of fish caught in November was richer in EPA and DHA than in the fish caught in June. The fish caught in November was also more unstable through frozen storage, due to a more unsaturated nature of the fatty acids present, indicating that special care needs to be applied during handling and treatment of golden redfish caught at this time. The light muscle had a higher nutritional value than the dark muscle and is a good nutritional source for human consumption. However, the dark muscle was prone to lipid oxidation which may have a negative influence on the more valuable light muscle. So there seems to be need to separate them.</p>
<p><i>English keywords:</i></p>	<p><i>Redfish; frozen storage; temperature fluctuation; quality deterioration</i></p>

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Introduction

Red fish

Redfish are important commercial species in the Icelandic fishing industry, in 2018 59.127 MT of redfish were caught. Redfish can be found all around Iceland, but the main fishing grounds are at the edge of the continental shelf south and west of Iceland at 200 to 400 m depth. It is caught mostly by trawlers throughout the year, but the largest catches are usually obtained in late winter and early spring (SAI Global, 2014). Redfish migrate between areas in the North Atlantic, related to oceanographic conditions, feeding and breeding areas (SAI Global, 2014).

Recently, redfish has become the second most commercially important fish species in Iceland. Redfish are processed into fresh, frozen and salted fillets. The main markets for redfish are Germany, Japan, China and South Korea (Icelandic Responsible Fisheries, 2016).

Effects of temperature abuse on fish quality

Freezing is one of the most common procedures applied to preserve physicochemical properties and to prolong storage life of fish products, especially fatty fish due to the high contents of unsaturated fatty acids present. The main purpose of freezing is therefore to prevent or slow down bacterial spoilage, enzyme activity and oxidation reactions.



Figure 1. Golden redfish (*Sebastes marinus*) (Jón Baldur Hlíðberg, www.fauna.is).

Quality changes of frozen fish during storage can be influenced by several factors including fish species, the biological status of fish at catch, handling on board, temperature and storage time before freezing, freezing rate, frozen storage temperature, temperature fluctuations, thawing procedure and protection from light and oxygen (Nielsen and Jørgensen, 2004). Optimal handling and transport conditions can be used to ensure high quality of the final fish

products, which arrive on the market (Ólafsdóttir, 2005). However, temperature fluctuations and abuse through the production and distribution chain can affect the fish quality and safety. These fluctuations mainly occur during handover from one party/function to the next in the value chain (Moureh and Derens, 2000). Studies have shown that unstable temperature accelerates the growth of specific spoilage organisms as well as pathogens (Rediers *et al.*, 2009), which can cause quality and safety problems and moreover economic losses. Furthermore, extracellular formation of ice crystals is accelerated during temperature fluctuations and hence cellular disruption is increased (Hagyard *et al.*, 1993; Bak *et al.*, 1999). Unstable temperature can therefore cause formation of large ice crystals within the fish muscle which have negative effects on its overall quality. Therefore, controlled temperature throughout the whole value chain is necessary to ensure product quality and stability. The effects of the temperature fluctuations and abuse on the product quality are depending on the temperature range. For example, fluctuations around -18 °C has proven to cause worse effects on the product quality than fluctuations around -25 °C since the freezing point of sodium chloride is -21.1 °C. For sensitive food products, such as fish, even short periods of temperature abuse can lead to significant loss of quality (WFLO, 2008). Temperature fluctuations can lead to an increase in the amount of unfrozen water in the product and changes in the structure of ice crystals and recrystallization. Thus, physicochemical deterioration increases as a result of enzymatic activity, lipid oxidation and breakdown of the physical structure (Nesvadba, 2008; Benjakul and Bauer, 2001; Karlsdóttir *et al.*, 2014; Zaritzky, 2008).

Effect of packing on fish quality

Packaging of fish generally consists of an outer layer (totes, cardboard cartons, waxed cartons or polystyrene) and an inner layer (plastic liners, bags or wraps) (Balasubramaniam & Chinnan, 1997; Kolbe & Kramer, 2007). Good packaging protects products against weight loss, flavor losses, nutritional losses, textural changes, formation of off odors and contamination with bacteria or other adulterants and physical damage (Kolbe & Kramer, 2007; Ebnesajjad, 2013).

The most suitable packaging material has a low rate of transmission of water vapor to avoid desiccation and a low permeability to oxygen to prevent fat and pigment oxidation. The material should be strong and tight fitting to prevent loss of moisture from the product, as

well as non-absorbing of oil or water, relatively inexpensive, easy to apply and easy to label (Kolbe & Kramer, 2007; Ebnesajjad, 2013).

Depending on the product and freezing method, packaging is either done before or after freezing. Fish blocks are packed in plastic bags, the frozen or arranged in trays to freeze and the packed. Layer packs are put in trays separated by plastic sheets and the frozen. After freezing, blocks without bags are packed in plastic bags, then placed in carton boxes. Layer packs are packed in waxed carton boxes (Balasubramaniam & Chinnan, 1997).

Changes in muscle pH and total volatile basic nitrogen

Post mortem changes in pH are caused by ATP hydrolysis. Muscle pH can also be affected by processing. The pH of fish fillets, which are treated by additives, especially phosphates, increases due to the use of alkaline phosphates that have strong anionic properties (Ünal *et al.*, 2004; Long *et al.*, 2011).

Total volatile basic nitrogen (TVB-N) is an important indicator used to assess the quality of fresh seafood products (Wu & Bechtel, 2008; Amegovu *et al.*, 2012). TVB-N contains mainly ammonia (NH₃), dimethylamine (DMA) and trimethylamine (TMA) which originate from bacterial and enzymatic decomposition of trimethylamine oxide (TMAO) (Castro *et al.*, 2006). The TMAO constitutes a characteristic and important part of the non-protein nitrogen fraction in marine species, which is responsible for osmoregulation in marine fish. After the death of fish, TMAO decreases due to an increase in bacterial and enzymatic activity (Ali *et al.*, 2010). TMAO is broken down into DMA and formaldehyde by TMAO dimethylase. In frozen fish, the TVB-N content, in the form of DMA and amino acids from proteins, increases with spoilage by enzymatic degradation (Ali *et al.*, 2010). However, in fresh fish, TMA is the main microbiological degradation product.

Thawing loss, water holding capacity and cooking yield

Freezing (Reid, 1997), frozen storage (Erickson, 1997; Sista *et al.*, 1997) and thawing methods (Roiha *et al.*, 2017) influence the quality of the product. The ability of the fish muscle to retain its natural quality is one of the main quality aspects both from the perspective of producers and customers (Olsson *et al.*, 2003).

The aim of the study was twofold. Firstly, to explore the influence of time and temperature during frozen storage on lipid deterioration of red fish by comparing the effect of temperature fluctuation and abuse during frozen storage, as can be expected during transportation. This was done by exploring these effects on the physicochemical characteristics and lipid stability of redfish fillets. Secondly, to investigate the effect of 4 days postcatch and 9 days postcatch, and seasonal variation on the quality and storage stability of frozen red fish.

This study forms part of PhD. research by Houg Thi Thu Dang and the results have been published in peer reviewed scientific papers. Hence, only the most relevant results and observation are summarized here. More detailed results can be observed in the before mentioned papers (Dang *et al.*, 2017 and Dang *et al.*, 2018).

Materials and methods

Raw material and experimental design

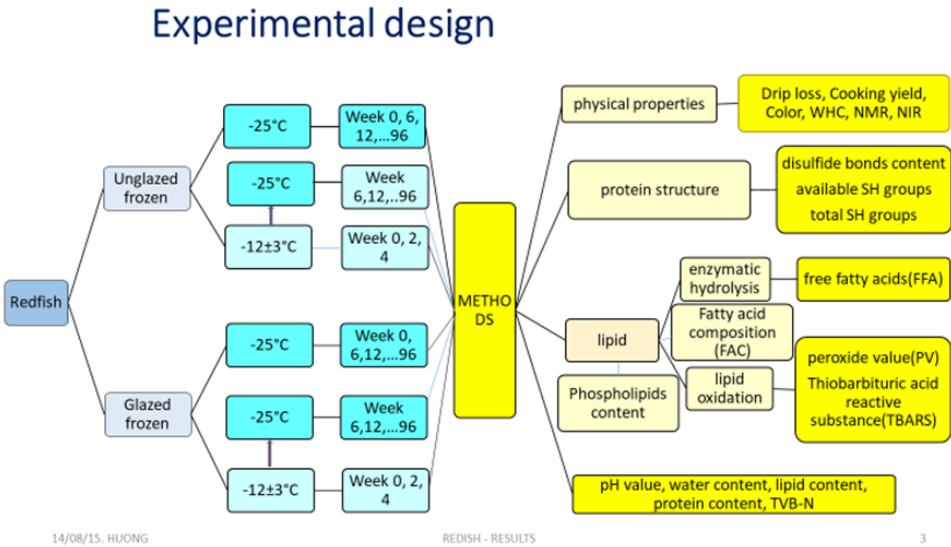


Figure 2. Experimental design for the project.

Table 1. Overview of the frozen redfish samplings (IQF=individually quick frozen).

Group*	Age of the raw materials before processing	Freezing method	Glazing	Storage** (°C)	Number of samplings
A	4 days	IQF	5%	-18 / -25	13
B	4 days	Layered	-	-18 / -25	13
C	9 days	IQF	5%	-18 / -25	13
D	9 days	Layered	-	-18 / -25	13
E	4 days	IQF	10%	-18 / -25	13

*Groups A-D sampled in June 2015 and November 2015. Group E sampled in November 2015.

**Storage in a freezing simulator at Matis.

Table 2. Overview of the groups for each catch.

Group	January 2015 ⁺	June 2015	November 2015	January 2016
A	x	X	X	X
B		X	X	X
C	x	X	X	X
D		X	X	X
E			X	

*Comparison of IQF fillets with and without glazing and comparison of stabilized and unstabilized temperature.

Raw materials, processing and sampling (January 2015)

Icelandic golden redfish (*S. marinus*) were caught off the southwest coast of Iceland in January 2015 by fresh fish trawlers. After landing, 4 days postcatch, the fish were transported to the factory for processing after 4 days postcatch. All samples were processed as skinless fillets, individually quick frozen, with or without glazing (5%). The fish was stored at stable condition for 24 months (-25°C) and unstable condition, first for 1 month (-25°C) then at -12°C at 1 month, and then at -25°C at the rest of the storage time. The fish was analyzed after 6, 9, 12, 15, 18, 21, and 24 months.

Raw materials, processing and sampling (June and November 2015)

The results from June and November were taken from the paper of Hung Thi Thu Dang *et al* (2018). Stability of Golden redfish (*Sebastian marinus*) during frozen storage as affected by raw material freshness and season of capture. Food Science and Nutrition.

Icelandic golden redfish (*S. marinus*) were caught off the southwest coast of Iceland in June and November 2015 by fresh fish trawlers. After landing, 4 days postcatch, the fish were processed and divided into two treatments, one processed 4 days postcatch and the other 9 days postcatch. All samples were processed as skinless fillets, frozen in layers separated in 4 layers

by polyethylene sheets, in a plate freezer, and were then packed in waxed cardboard boxes, and finally stored at -25°C.

Analyses were performed after 0, 4, 8, 12, 16, and 20 months of frozen storage at -25°C. Prior to analysis, one pack from each treatment was thawed at 4°C for at least 24 hr. After thawing, fish from each treatment were divided into three groups, each containing ten fillets (average weight per fillet was 120 ± 20 g). Light and dark muscles of the fillet were manually and used for all analyses.

Physical and chemical analysis (January, June and November 2015)

Water, total lipids and phospholipid content

Water content was determined by drying 5 g of minced sample at 102-104 °C for 4 h (ISO, 1999). The results were calculated as the weight loss during drying as a percentage of the wet muscle.

Total lipids (TL) of the samples were extracted from 25 g of samples (the weight was adjusted according to the water content of each sample) with methanol/chloroform/0.88% KCl (at 1/1/0.5, v/v/v) according to the method of Bligh and Dyer (1959). The lipid content was determined gravimetrically, and results were expressed as a percentage of the wet muscle.

Phospholipids (PL) content of the fish muscles was determined on the TL extracts by using a colorimetric method (Stewart, 1980). Phosphatidylcholine in chloroform (1 mg/mL) was prepared for a standard curve, and results were expressed as g phospholipids/100 g TL.

Water activity (a_w)

Water activity (a_w) is the partial vapor pressure of pure water at the same temperature. Pure distilled water has a water activity of exactly one. Higher a_w substances tend to support more microorganisms (Karel *et al.*, 1975). The samples were measured with Dew point water activity meter from AQUALAB.

Fatty acid composition

The fatty acid composition of the sample was determined on the TL extracts by gas chromatography (Varian 3900 GC, Varian, Inc., Walnut Creek, CA, USA). The methylation of

fatty acids was carried out according to the AOAC Ce 1b-89 (1998) method. The programme was based on the AOAC 996.06 (2001) method. Results were expressed as a percentage of TL.

Free fatty acid content

Free fatty acids (FFA) content was determined by the Lowry and Tinsley (1976) method with modifications described by Bernardez *et al.* (2005). The absorbance of the solution was read at 710 nm (UV-1800 spectrophotometer, Shimadzu, Japan) and compared to a standard curve prepared from oleic acid in a concentration range of 2–14 μmol . Results were expressed as g FFA per 100 g TL.

Lipid oxidation

Lipid hydroperoxides (PV), a primary oxidation product, was determined by the ferric thiocyanate method (Shantha & Decker, 1994) with modifications according to Romotowska *et al.* (2016), except that the lower chloroform layer containing lipids was collected (0.2 mL for the dark and 0.5 mL for the light muscle) and mixed with 0.8 and 0.5 mL of the chloroform:methanol (1:1) solution for the dark and light muscle, respectively. The results were expressed as μmol lipid hydroperoxide per kg of the sample ($\mu\text{mol}/\text{kg}$ muscle).

Thiobarbituric acid reactive substances (TBARS), secondary oxidation products, were determined by the method of Lemon (1975) with modifications as described by Romotowska *et al.* (2016), except that an amount of 0.8 and 0.5 mL thiobarbituric acid (0.02 M) were mixed with 0.2 mL collected supernatant for the dark and 0.5 mL for the light muscle, respectively. The results were expressed as μmol malondialdehyde diethyl acetal per kg of wet muscle (μmol MDA/kg muscle).

Protein content

Protein content was determined according to ISO-5983-2 (2009) and expressed as a percentage of the wet muscle.

pH, thawing loss and cooking yield

The muscle pH was measured by inserting the pH probe (Radiometer PHM80 Portable pH meter, Denmark) directly into the muscle samples.

Thawing losses were calculated as the ratio (%) of liquid lost during thawing to the weight of the individual frozen blocks.

Cooking yield (CY) was calculated as the ratio (%) of the sample weight after cooking to the weight of the sample before cooking. About 35 g of each fillet (n = 5 from each group) were weighed and heated in a steaming oven (Convotherm OGS 6.10 Combi convection steam oven, Elektrogeräte GmbH, Eglfing, Germany) at 100 °C for 10 minutes. Samples were drained for 10 minutes prior to being weighed again.

Color

The color of the samples was determined with Minolta Chroma Meter CR-400 (Minolta, Osaka, Japan) using the CIE Lab system. The instrument recorded the *L*-value, indicating lightness on the scale from 0 to 100 from black to white, the *a*-value, ranging from (+) red to (-) green, and the *b*-value, ranging from (+) yellow to (-) blue. The color was measured at five positions above the lateral line, from the head to the tail of each fillet on the light (inner) side of the fillet.

Chemical analyses were performed separately on the light and dark muscle of each group. Water, total lipids (TL), phospholipids (PL), fatty acid composition, lipid oxidation and hydrolysis products, and pH were performed on both muscle types, while protein content, water holding capacity (WHC), and total volatile basic nitrogen (TVB-N) analysis were performed only on the light muscle. Analysis were set up in triplicate of muscle pH, PL content, peroxide values (PV), and thiobarbituric acid reactive substances (TBARS), while analyses for WHC, TVB-N, protein, water, TL content, free fatty acids (FFA), and fatty acid composition were performed in duplicates. Thawing loss, color and cooking yield (CY) analyses were performed on individual fillets. The weight proportion of muscle types (light and dark) of the fillets was performed at month 0 of storage for each fillet.

The weight proportions were calculated as the ratio (%) of the dark and light muscle weight compared to the weight of the whole fillet. Description of other chemical and physical analyses appears in chapter concerning “Raw materials, processing and sampling (January 2015).

Ice percentage

The ice percentage was measured by weighing the redfish when it was quick frozen and weighing again after storage.

Statistical analysis

Data summaries and statistical analyses were carried out and figures were drawn using the STATISTICA software (Version 10.0, StatSoft, OK 74104 USA), and Microsoft Office Excel 2013 (Microsoft Inc. Redmond, WA, USA). One-way ANOVA, Tukey HSD's test and Student t test for independent samples were performed on the means of each variable. Pearson correlation analysis was performed to find the correlations between variables. Significance of difference was defined at $p < 0.05$ for all statistical analyses. Principal components analysis (PCA) was performed using Unscrambler[®] (Version 10.2, CAMO ASA, Trondheim, Norway) to identify similarities and differences between samples. All variables were weighed with the inverse of the standard deviation to correct for different scales of the variables.

Results and discussion

Raw materials, processing and sampling (January 2015)

Fat content

Higher fat content were generally observed in glazed and unglazed dark muscle of redfish stored at -12°C and -25°C compared with glazed and unglazed white muscle (Figure 3 and 4). This is in agreement with the findings of Karlsdóttir *et al.* (2014) on the composition of saithe and hoki. The dark muscle was significantly higher in fat content compare with the white muscle. No differences were observed in the glazed and unglazed white muscle for both temperatures. But slightly differences were obeserved in the glazed and unglazed dark muscle for both storage temperatures.

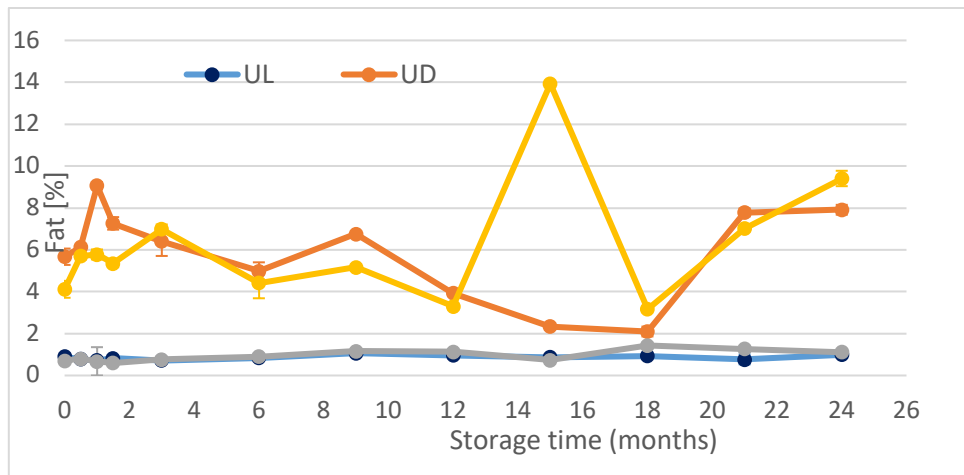


Figure 3. Fat content in unglazed light muscle (UL), unglazed dark muscle (UD), glazed light muscle (RL) and glazed dark muscle (RD) in redfish. Stored at -12°C.

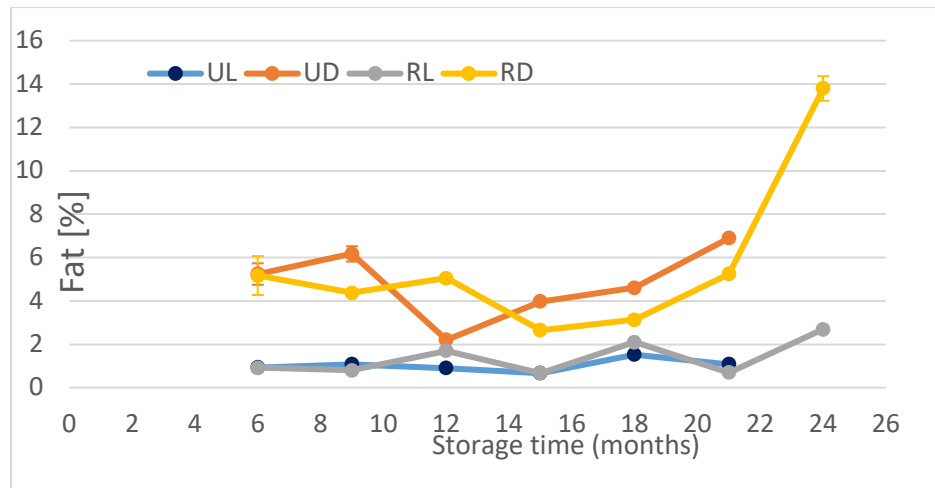


Figure 4. Fat content in unglazed light muscle (UL), unglazed dark muscle (UD), glazed light muscle (RL) and glazed dark muscle (RD) in redfish. Stored at -25°C.

Free fatty acid (FFA)

Lipid hydrolysis may occur postmortem in fish leading to an increase in FFA content, due to increased lipase and phospholipase activities (Pacheco-Aguilar *et al.*, 2000). Higher FFA content was observed in the glazed and unglazed light muscle, compared with the glazed and unglazed dark muscle in both storage time (-12°C and -25°C) (Figure 5 and 6).

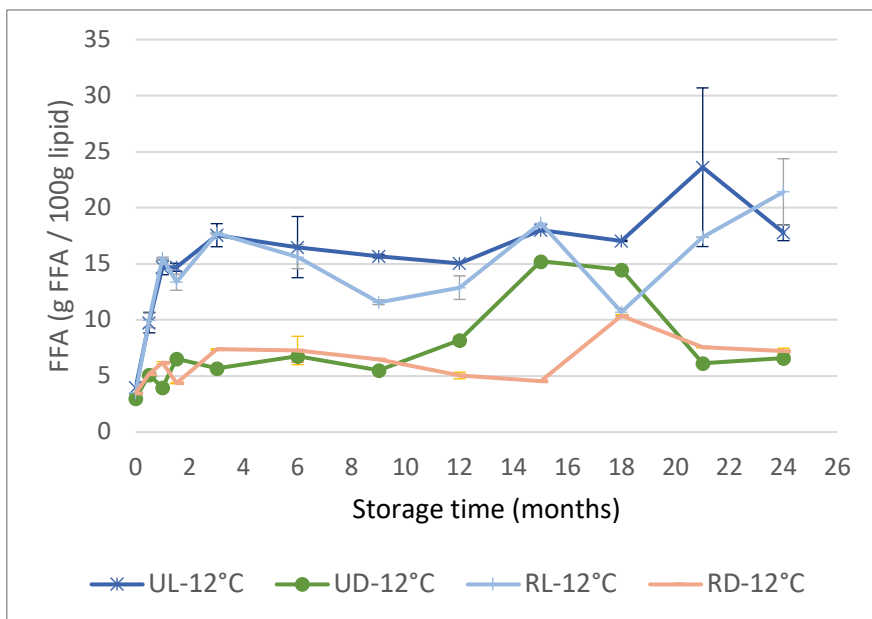


Figure 5. Free fatty acids (FFA) content in unglazed light muscle (UL), unglazed dark muscle (UD), glazed light muscle (RL) and glazed dark muscle (RD) in redfish, stored at -12°C.

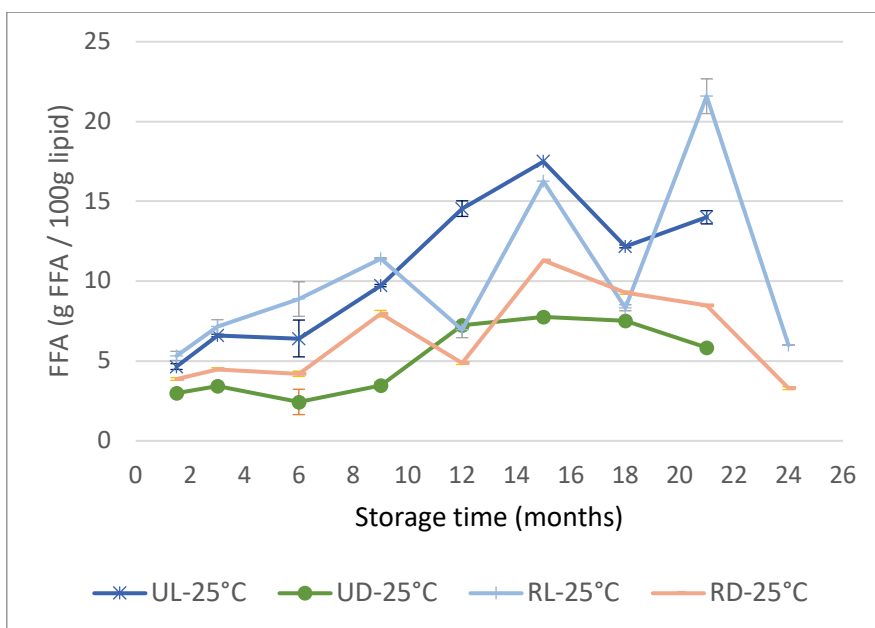


Figure 6. Free fatty acids (FFA) content in unglazed light muscle (UL), unglazed dark muscle (UD), glazed light muscle (RL) and glazed dark muscle (RD) in redfish, stored at -25°

Peroxide value (PV)

The peaks in PV were followed by decomposition of hydroperoxides to secondary oxidation products until from month 12 to 18 of frozen storage for the glazed and unglazed dark muscle for both storage temperatures, respectively (Figure 7 and 8). At this time range the PV value for unglazed dark muscle at -12°C storage was lower (480µmol/kg) than for the glazed dark

muscle (850 $\mu\text{mol/kg}$) (Figure 7). The PV values for the light muscle was rather low and stable through the storage time.

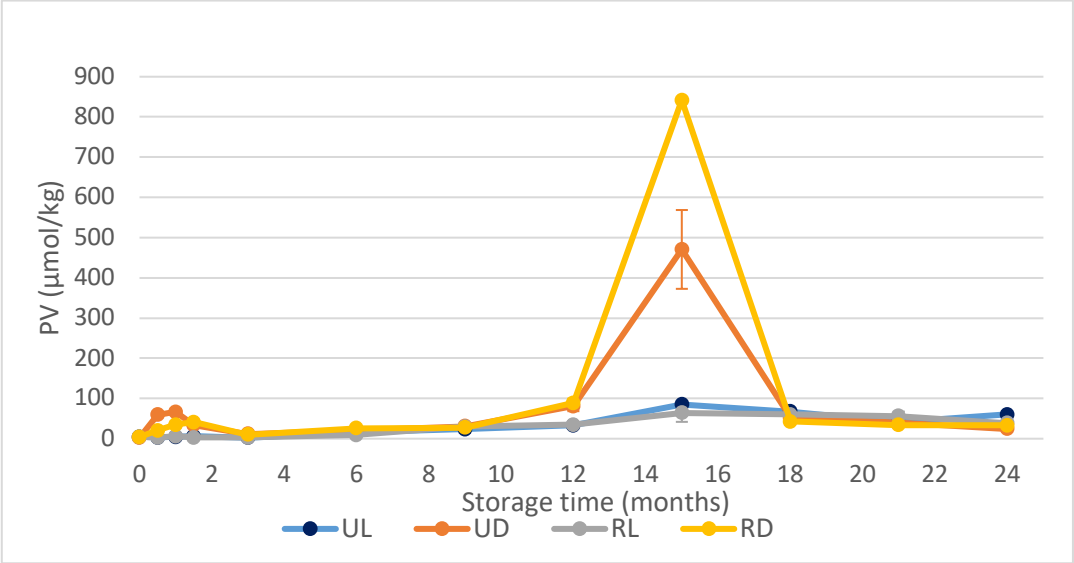


Figure 7. Peroxide value (PV) content in unglazed light muscle (UL), unglazed dark muscle (UD), glazed light muscle (RL) and glazed dark muscle (RD) in redfish, stored at -12°C.

The highest peak value for PV in samples stored at -25°C was 700 $\mu\text{mol/kg}$ for both glazed and unglazed dark muscle after 15 months (Figure 8). The light muscle was rather stable with low PV value.

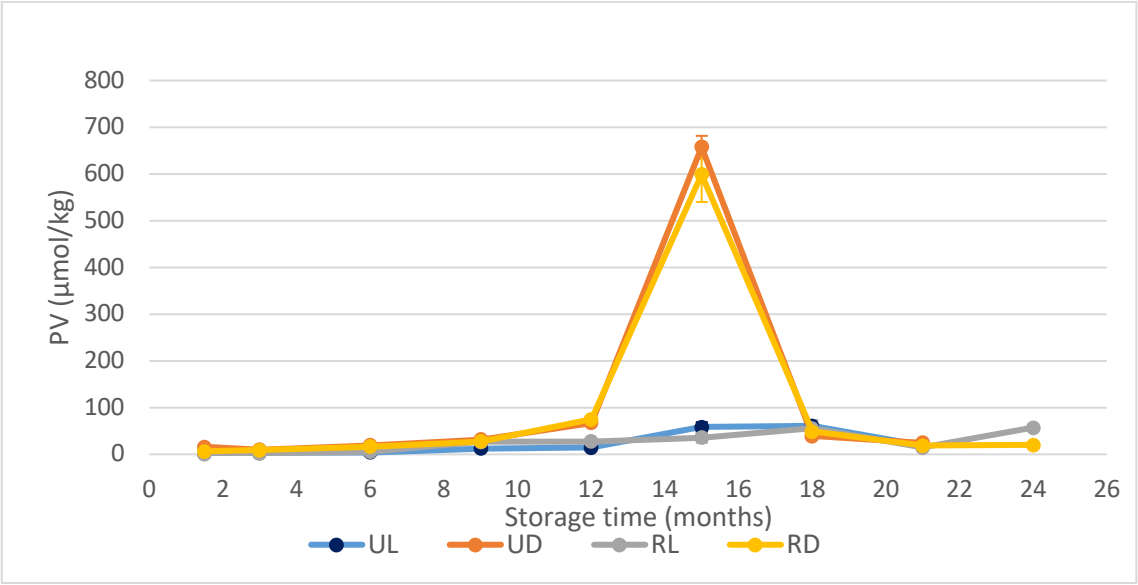


Figure 8. Peroxide value (PV) content in unglazed light muscle (UL), unglazed dark muscle (UD), glazed light muscle (RL) and glazed dark muscle (RD) in redfish, stored at -25°C.

Thiobarbituric acid reactive substances (TBARS)

The TBARS content was observed at the same storage points as the PV values for the light muscles. The TBARS for the dark muscle fluctuated during the storage time for both storage temperatures (-12 °C and -25°C) (Figure 9 and 10). The TBARS in the dark muscle stored at -25°C tended to increase during the storage time (Figure 10).

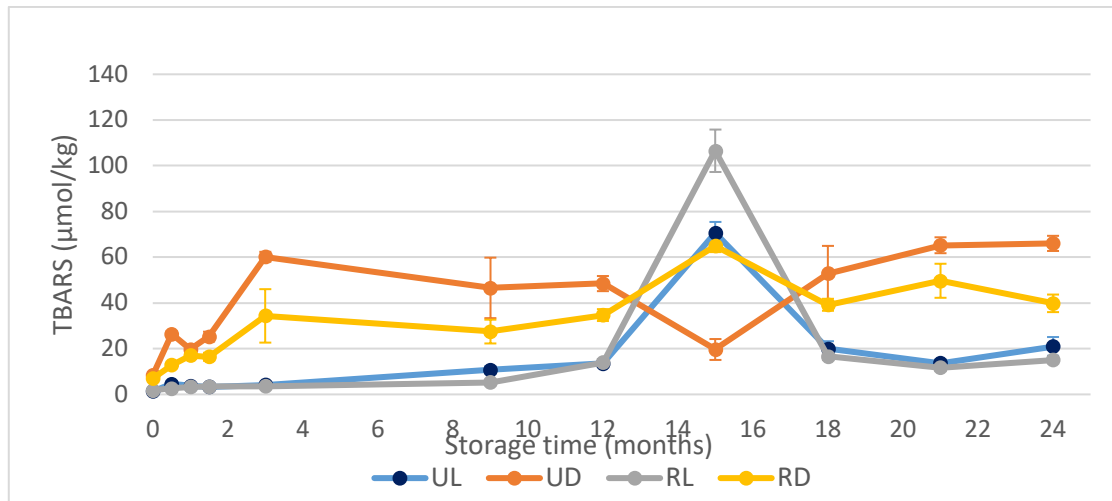


Figure 9. Thiobarbituric acid reactive substances (TBARS) content in unglazed light muscle (UL), unglazed dark muscle (UD), glazed light muscle (RL) and glazed dark muscle (RD) in redfish. Stored at -12°C.

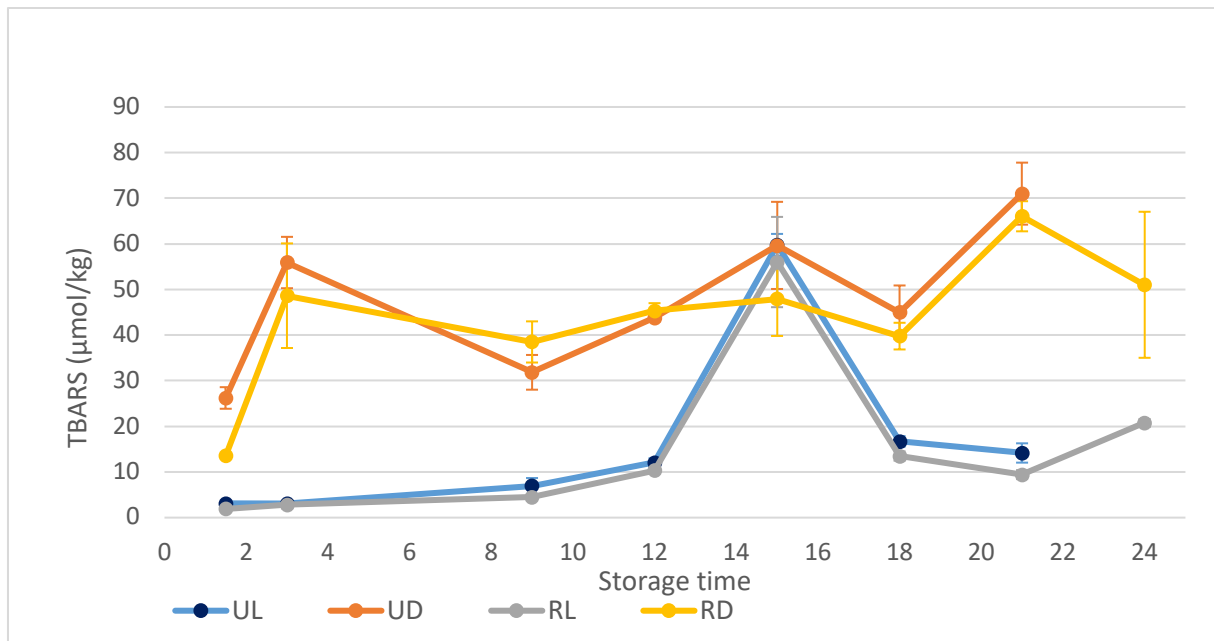


Figure 10. Thiobarbituric acid reactive substances (TBARS) content in unglazed light muscle (UL), unglazed dark muscle (UD), glazed light muscle (RL) and glazed dark muscle (RD) in redfish. Stored at -25°C.

Cooking yield

Cooking yield in glazed and unglazed fillets of redfish for both storage temperatures was stable during the 24 months of storage with a 75% yield (Figure 11 and 12).

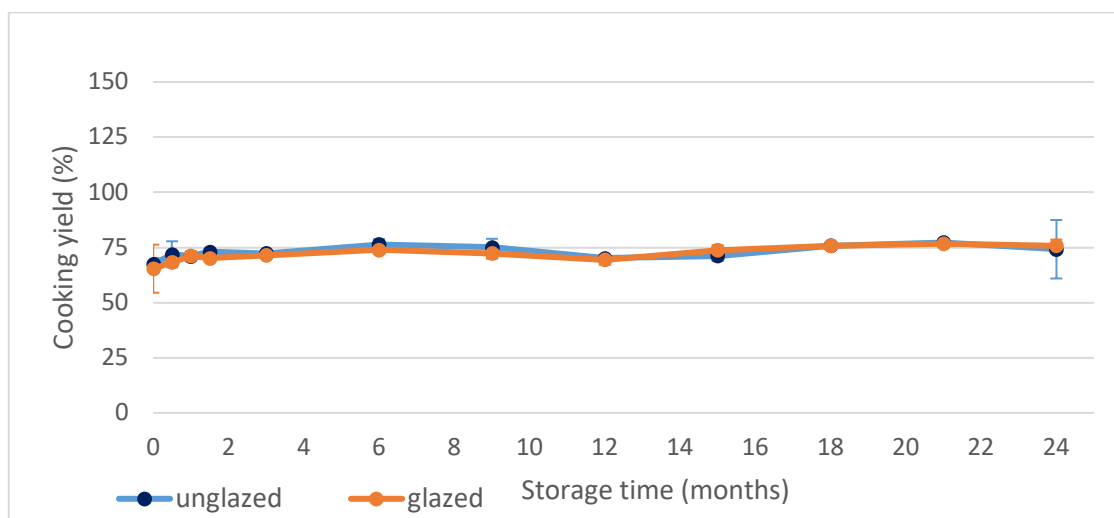


Figure 11. Cooking yield (%) in glazed and unglazed redfish, stored at -12°C.

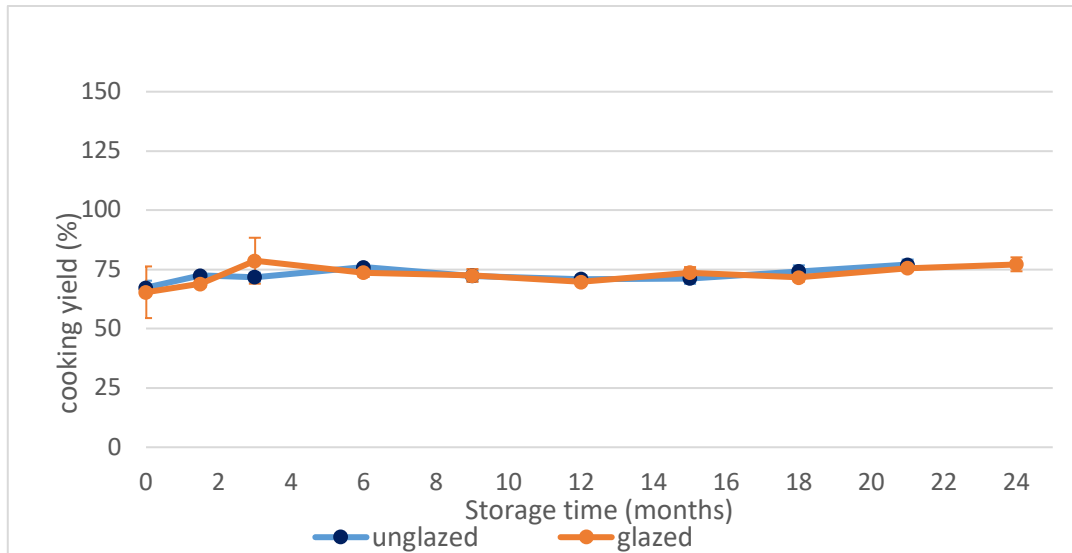


Figure 12. Cooking yield (%) in glazed and unglazed redfish, stored at -25°C.

Color

The lightness (*L*-value) of the fillets (stomach and the back) was stable during the storage period, both for glazed and unglazed fillets stored at -12°C (Figure 13), with values between 50 and 60.

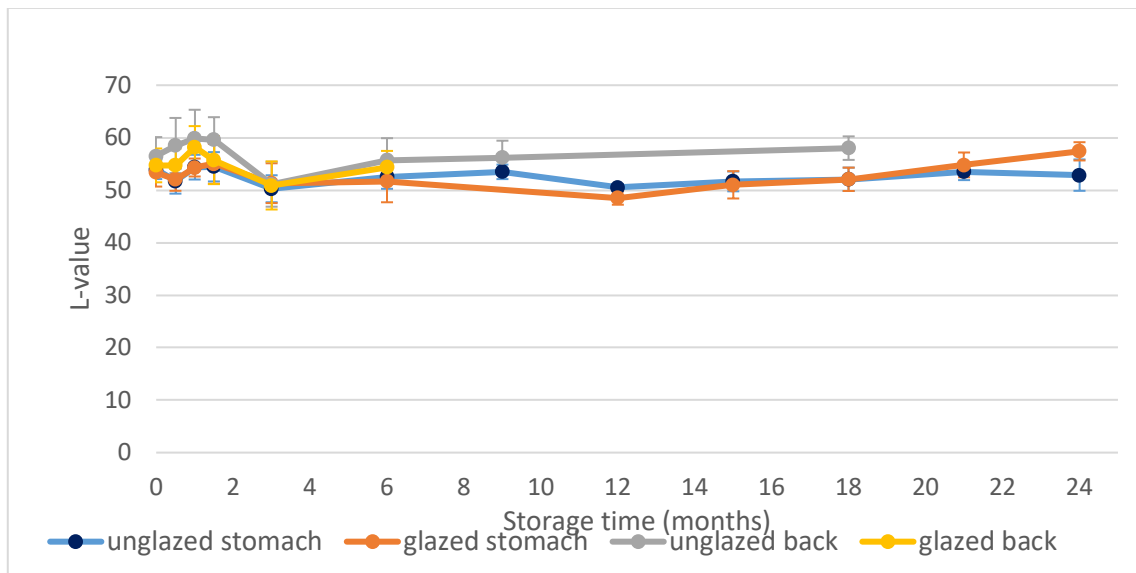


Figure 13. *L*-value (dark-light, 0-100) in unglazed and glazed stomach, and glazed and unglazed back of red fish. Stored at -12°C.

The *a*-value of the fillets, stored at -12°C, fluctuated slightly in the beginning of the storage, both for stomach and back fillets. In general the *a*-values of the fillets were stable (Figure 14)

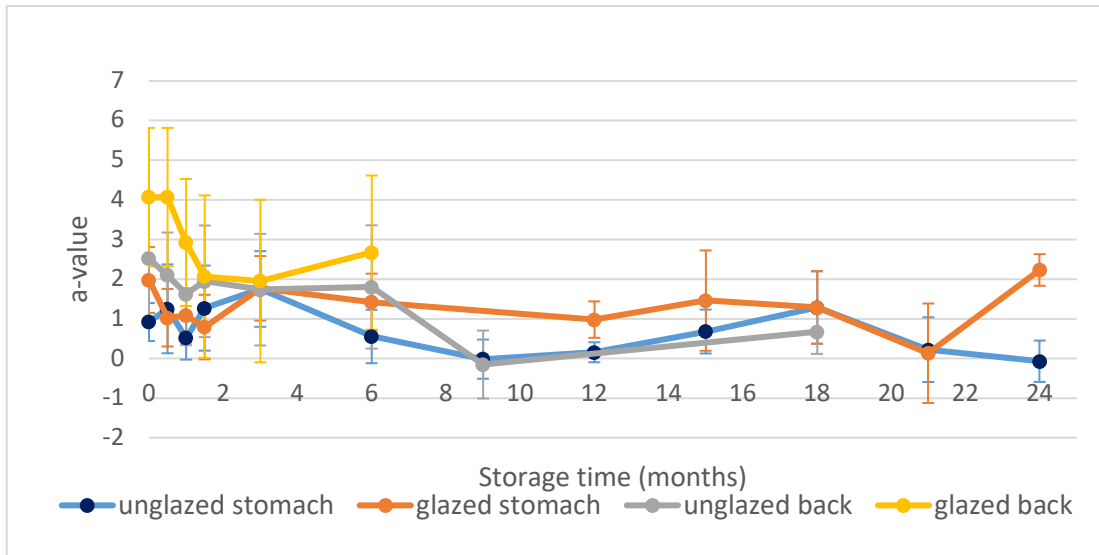


Figure 14. *a*-value ranging from (+) red to (-) green, in unglazed and glazed stomach, and glazed and unglazed back of red fish. Stored at -12°C.

The *b*-value fluctuated slightly for all the fillets stored at -12°C, in the beginning of the storage period, but after 6 months the *b*-value was rather stable (Fig 15).

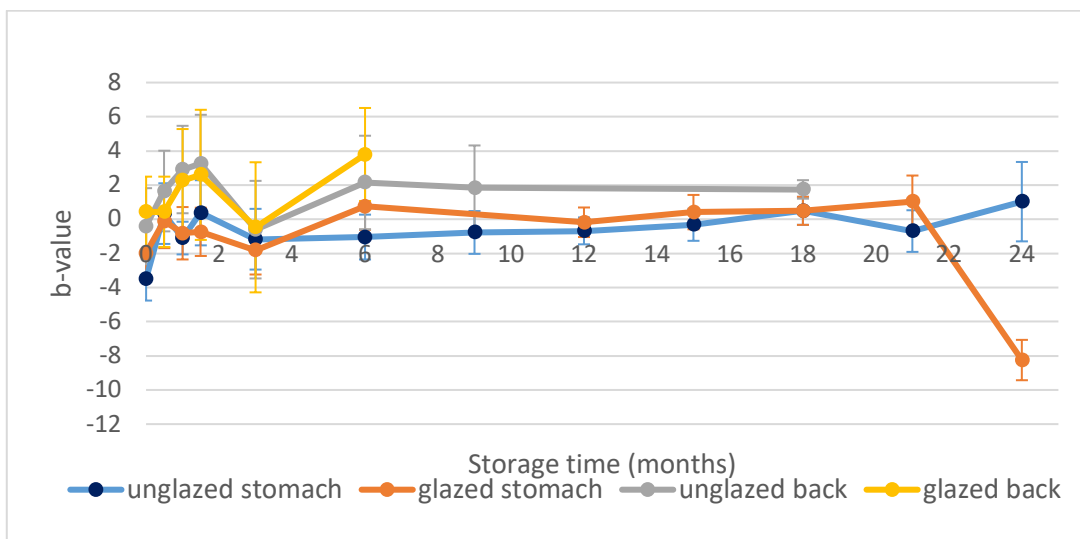


Figure 15. *b*-value, ranging from (+) yellow to (-) blue, in unglazed and glazed stomach, and glazed and unglazed back of red fish. Stored at -12°C.

The lightness (*L*-value) of the fillets (stomach and back) stored at -25°C was stable through the storage time for 24 months, with a *L*-value around 50 (Figure 16).

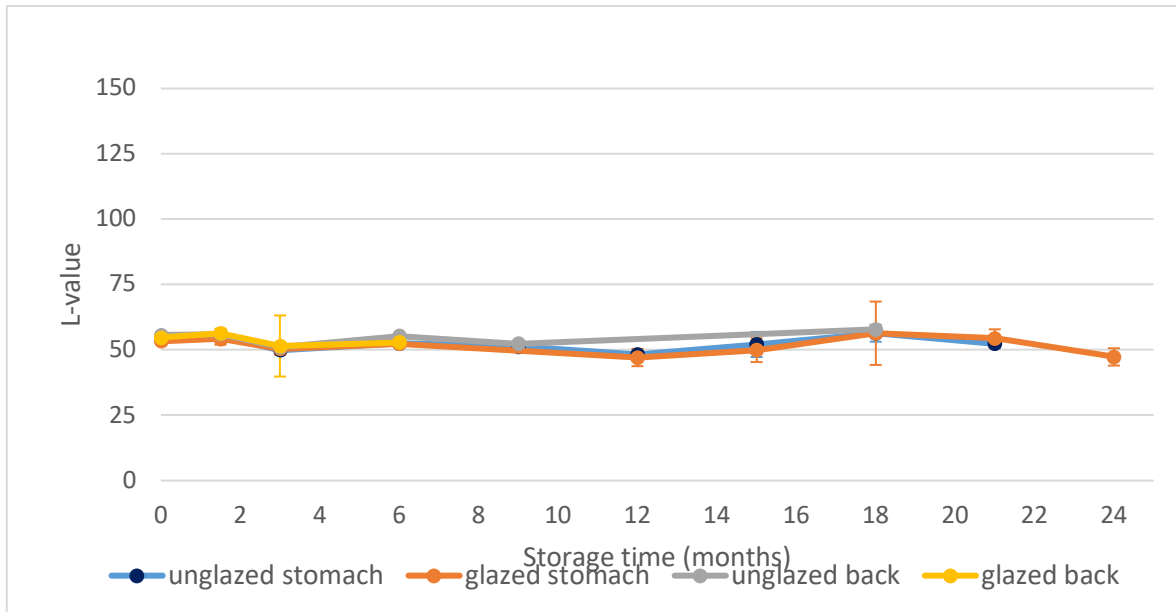


Figure 16. L-value (dark-light, 0-100) in unglazed and glazed stomach, and glazed and unglazed back of red fish, stored at -25°C.

For the first 6 months the a -value fluctuated slightly in fillets stored at -25°C, but after that the a -value was rather stable (Figure 17).

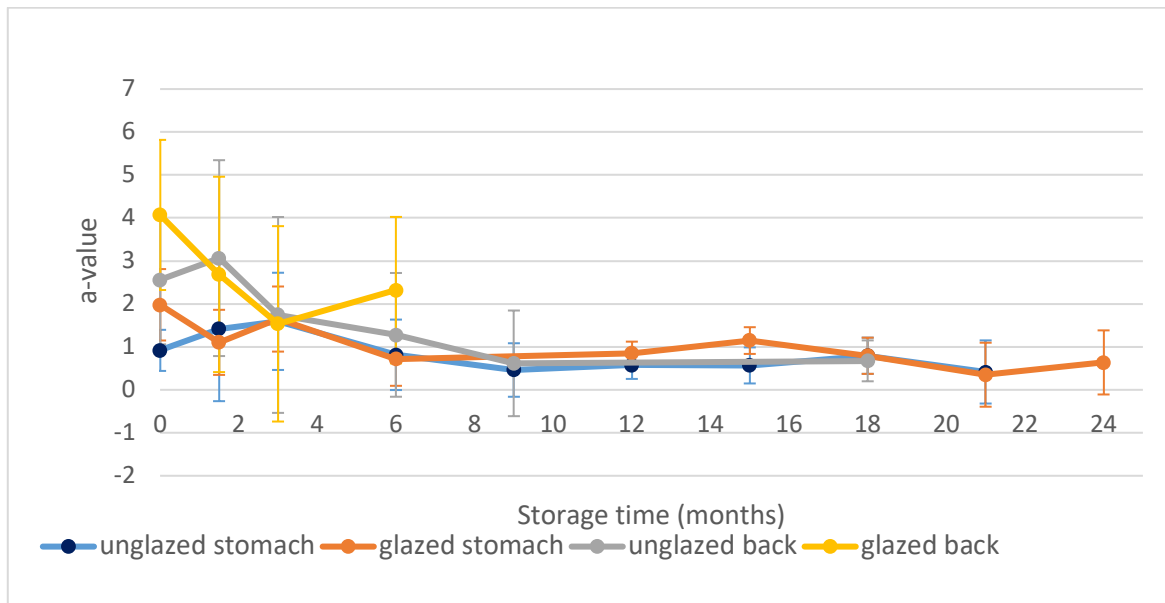


Figure 17. a -value ranging from (+) red to (-) green, in unglazed and glazed stomach, and glazed and unglazed back of red fish. Stored at -25°C.

Also the b -value fluctuated for the first 6 months in fillets stored at -25°C, but after that the b -value for both treatments were stable (Figure 18).

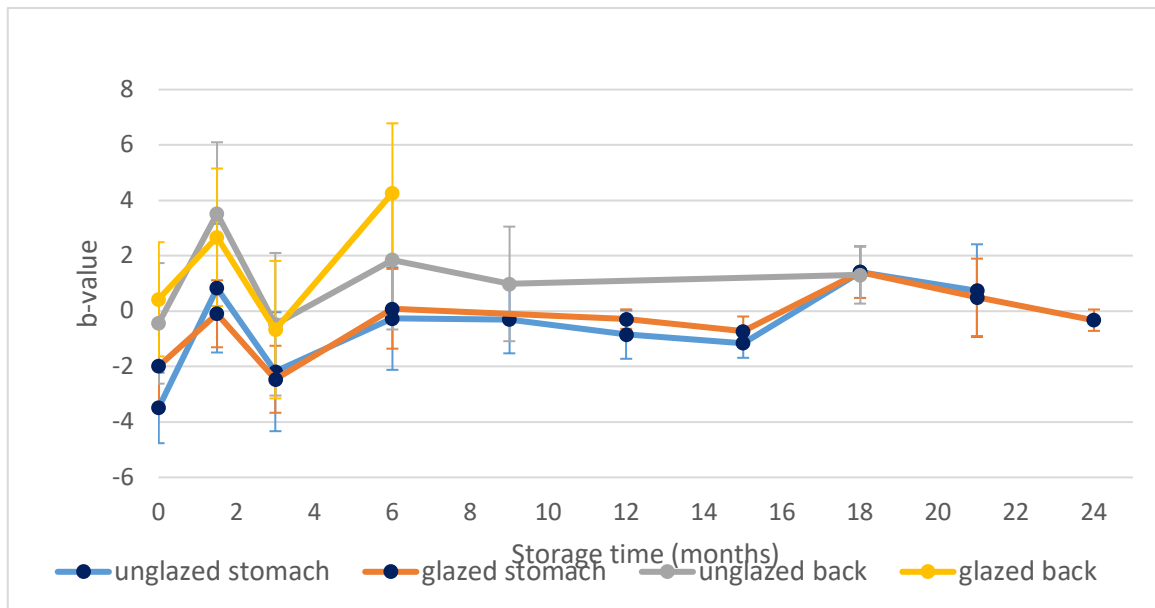


Figure 18. b-value, ranging from (+) yellow to (-) blue, in unglazed and glazed stomach, and glazed and unglazed back of red fish. Stored at -25°C.

The lightness of glazed and unglazed light and dark muscle stored at -12°C was stable during the storage time (Figure 19). During the first three months of storage the lightness fluctuated slightly.

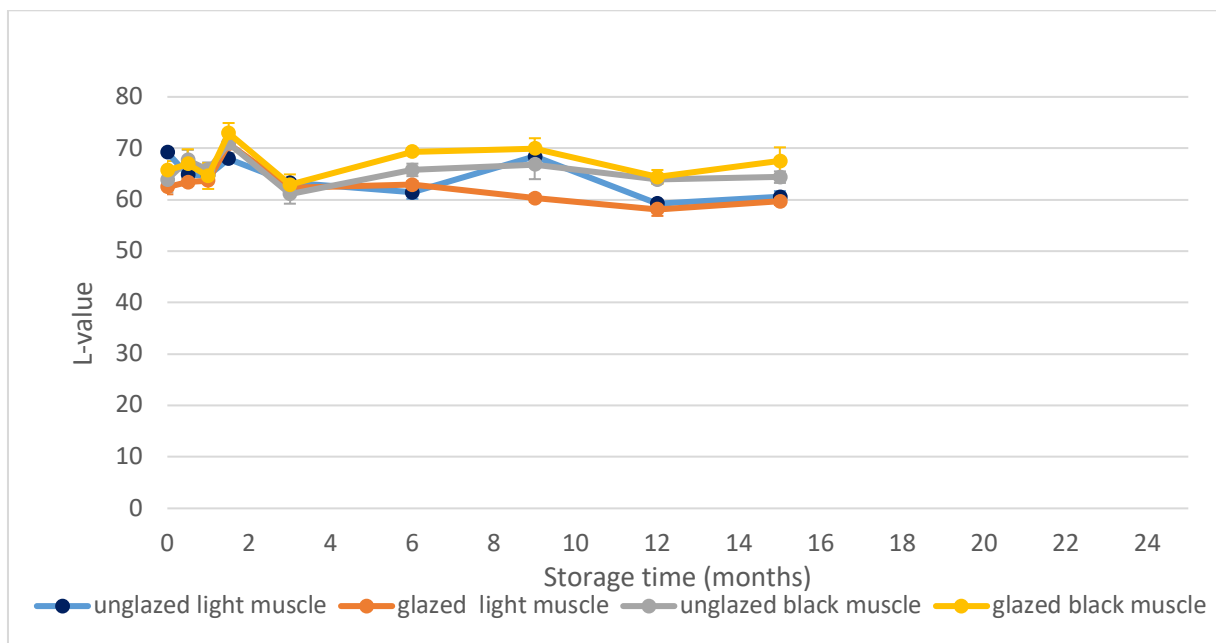


Figure 19. L-value (dark-light, 0-100) in unglazed and glazed light muscle, and glazed and unglazed dark muscle of red fish, stored at -12°C.

The glazed and unglazed dark muscle had higher a -value (more red) than the glazed and unglazed light muscle stored at -12°C (Figure 20). The a -values decreased for both the dark and light muscles during the storage period.

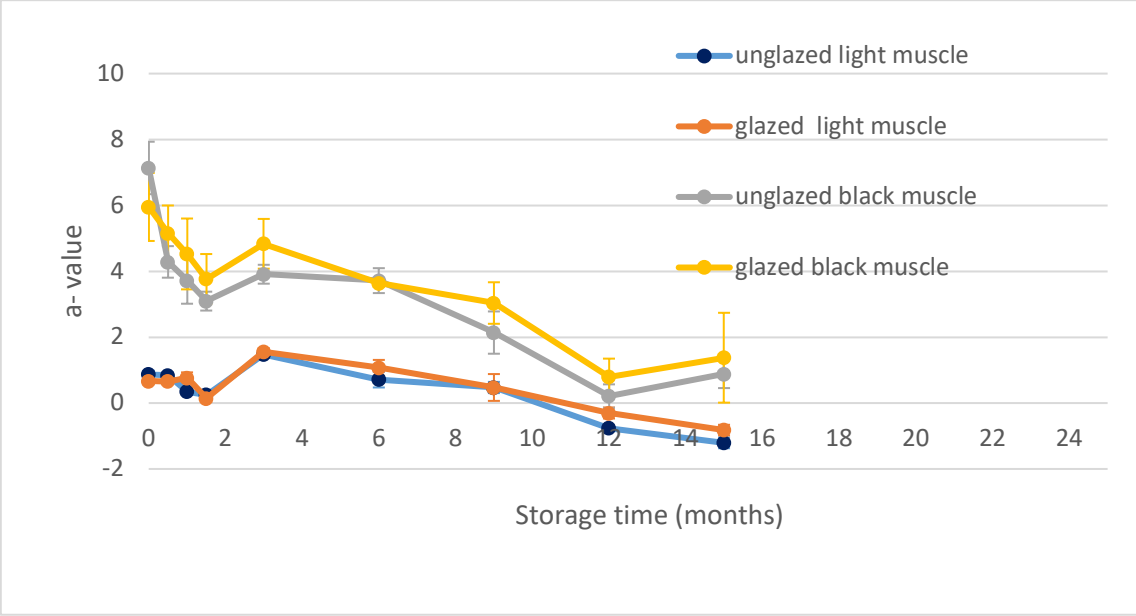


Figure 20. a -value ranging from (+) red to (-) green, in unglazed and glazed light muscle, and glazed and unglazed dark muscle of red fish. Stored at -12°C .

Significantly higher b -values were observed in the dark muscle stored at -12°C compared with the light muscle (Figure 21). The b -values fluctuated slightly for both the light and dark muscles.

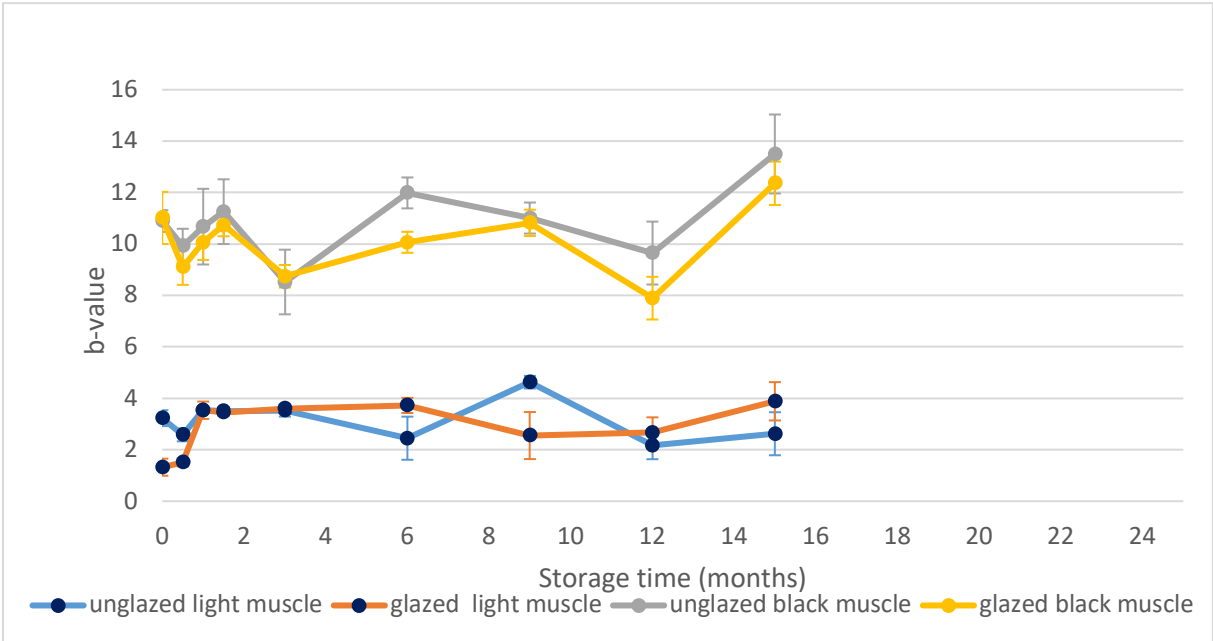


Figure 21. b -value, ranging from (+) yellow to (-) blue, in unglazed and glazed light muscle, and glazed and unglazed dark muscle of red fish, stored at -12°C .

No differences were found in L -values between the light and the dark muscles in redfish stored at -25°C (Figure 22). The L -values were rather stable through the storage period for both types of muscle.

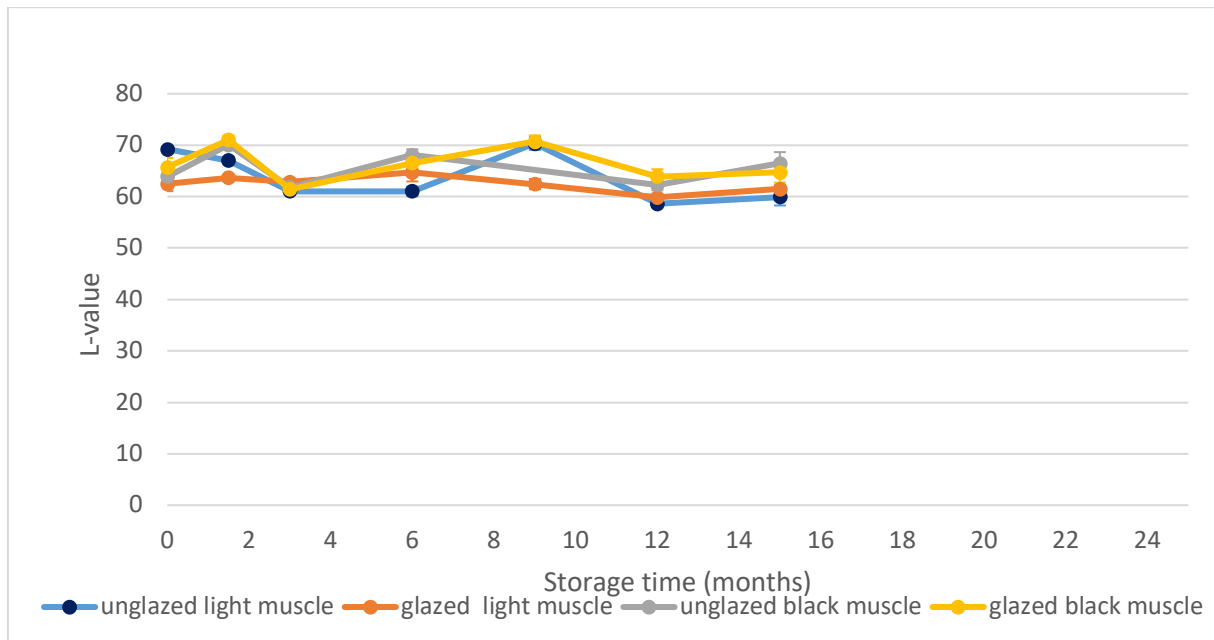


Figure 22. L -value (dark-light, 0-100) in unglazed and glazed light muscle, and glazed and unglazed dark muscle of red fish, stored at -25°C .

The a -values in dark and light muscle in fillets decreased during the storage period of redfish stored at -25°C (Figure 23). The a -value in the dark muscle was significantly higher in the dark muscle compared with the light muscle for the first 12 months. From month 12 to month 15 the a -values were identical for both types of muscles.

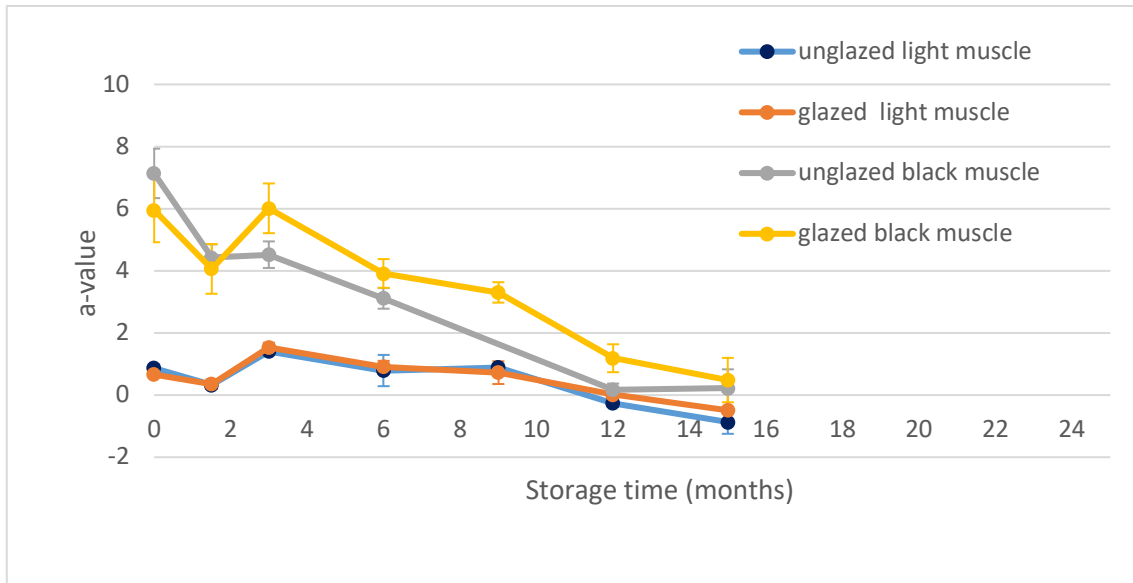


Figure 23. *a*-value ranging from (+) red to (-) green, in unglazed and glazed light muscle, and glazed and unglazed dark muscle of red fish. Stored at -25°C.

The *b*-value was significantly higher in the dark muscle, compared with the light muscle for both treatments of redfish stored at -25°C (Figure 24). The *b*-value fluctuated slightly during the storage period

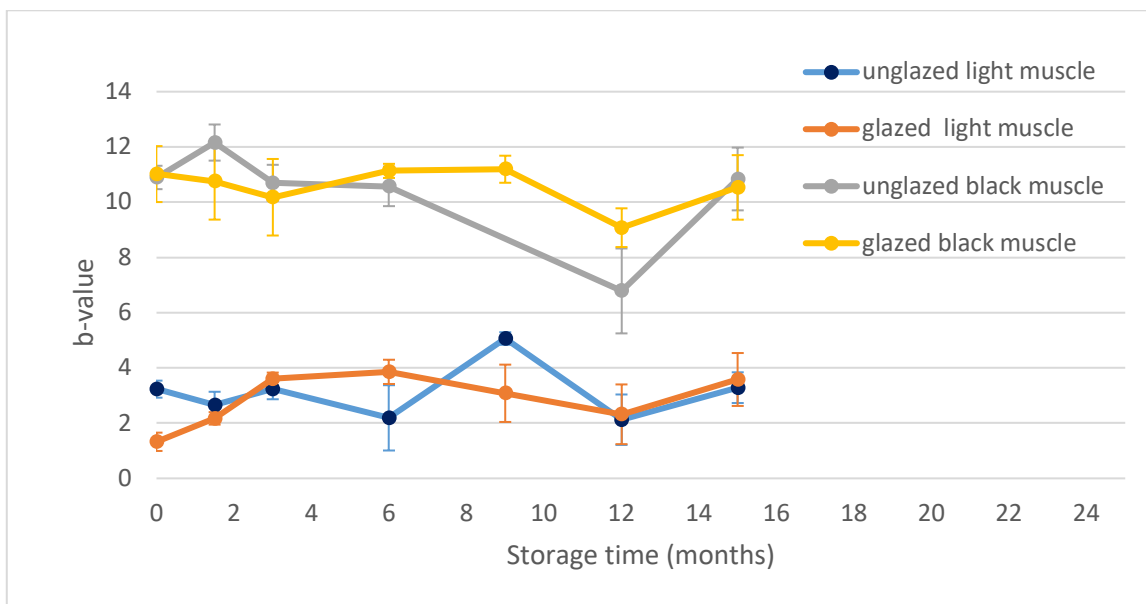


Figure 24. *b*-value, ranging from (+) yellow to (-) blue, in unglazed and glazed light muscle, and glazed and unglazed dark muscle of red fish, stored at -25°C.

Water content

The water content in glazed and unglazed light muscle was significantly higher compared with the glazed and unglazed dark muscle stored at -12°C (Figure 25). Glazing had no affect on the

water content. The light muscle was more stable in water content than the dark muscle, were the latter fluctuated during the storage time.

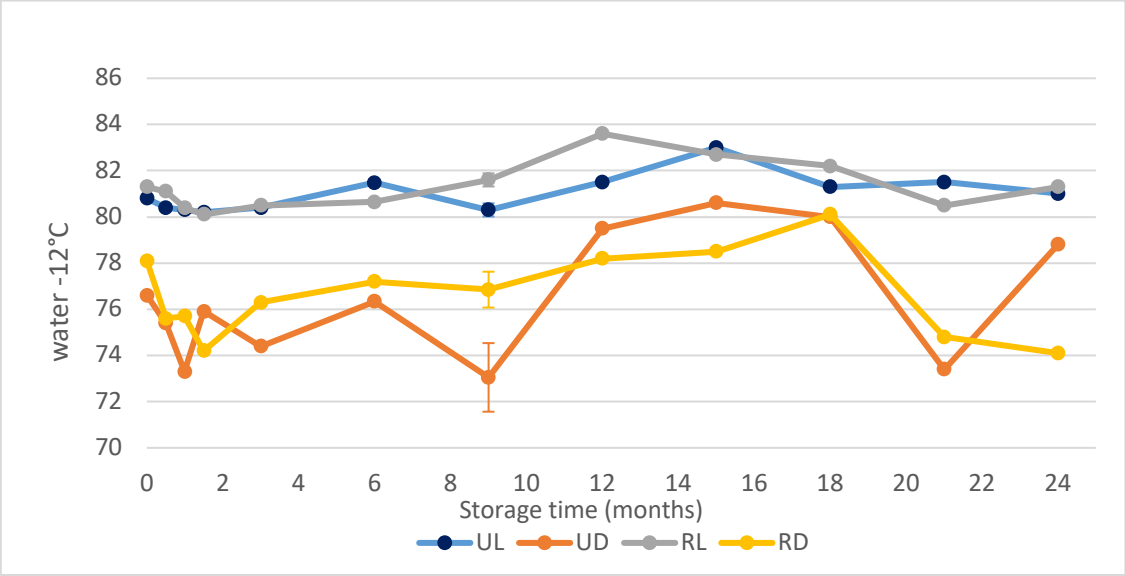


Figure 25. Water content in UL: unglazed light muscle; RL: glazed light muscle; UD: unglazed dark muscle and RD: glazed dark muscle in redfish, stored at -12°C.

The water content was significantly higher in the light muscle, compared with the dark muscle, for both treatments in redfish stored at -25°C (Figure 26). The water content in the light muscle was more stable, while the water content in the dark muscle fluctuated. After 18 months the water content decreased for both type of muscles.

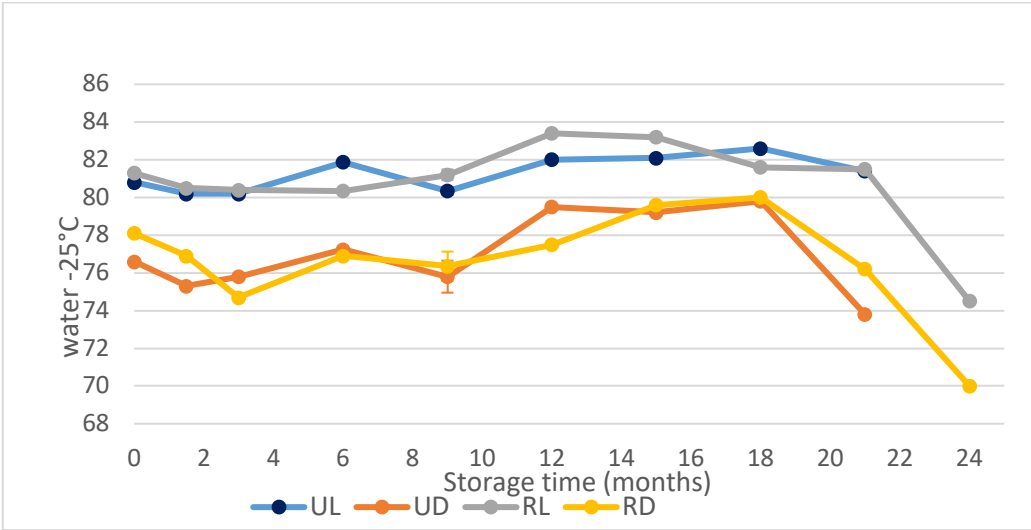


Figure 26. Water content in UL: unglazed light muscle; RL: glazed light muscle; UD: unglazed dark muscle and RD: glazed dark muscle in redfish, stored at -25°C.

Phospholipids (PL)

Phospholipids content was higher in the light muscle compared with the dark muscle during most of the storage period of redfish stored at -12°C (Figure 27). This is in agreement with previous studies of saithe and hoki (Karlsdóttir *et al.*, 2014) and herring (Dang *et al.*, 2017). PL content after 9 months of storage increased in the light muscle, while after in the dark muscle the PL content increased after 12 months. PL content decreased for the remaining duration of storage. This decrease in PL content for both type of muscles was expected due to the hydrolytic activities by phospholipases (Sista *et al.*, 1997).

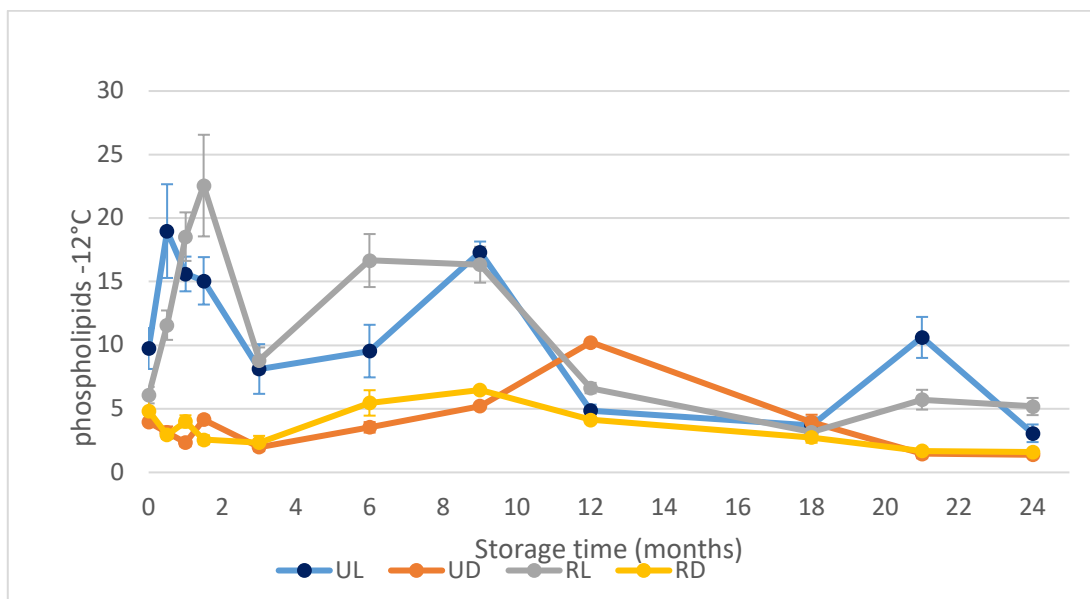


Figure 27. Phospholipid content in UL: unglazed light muscle; RL: glazed light muscle; UD: unglazed dark muscle and RD: glazed dark muscle in redfish, stored at -12°C .

PL content increased in light muscle during storage of 9 months in redfish stored at -25°C (Figure 28). Also the PL content in the light muscle was significantly higher, during storage of 9 months, compared with the dark muscle. The PL content in the light muscle fluctuated during storage time, while the PL content in the dark muscle remained rather stable. PL content decreased for the remaining duration of storage. PL content in the dark muscle increased during storage of 12 months and decreased for the remaining duration of storage.

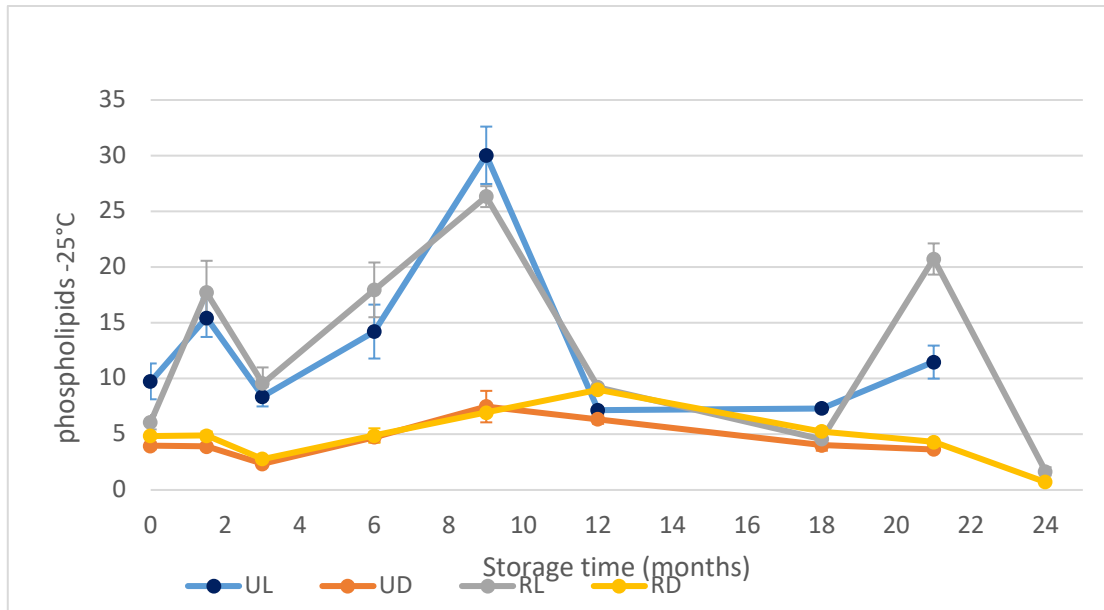


Figure 28. Phospholipid content in UL: unglazed light muscle; RL: glazed light muscle; UD: unglazed dark muscle and RD: glazed dark muscle in redfish, stored at -25°C.

pH

The pH of the light muscle in redfish stored at -12°C was between 6.4 -6.8, while in the dark muscle the pH was between 6.4 – 6.7 (Figure 29).

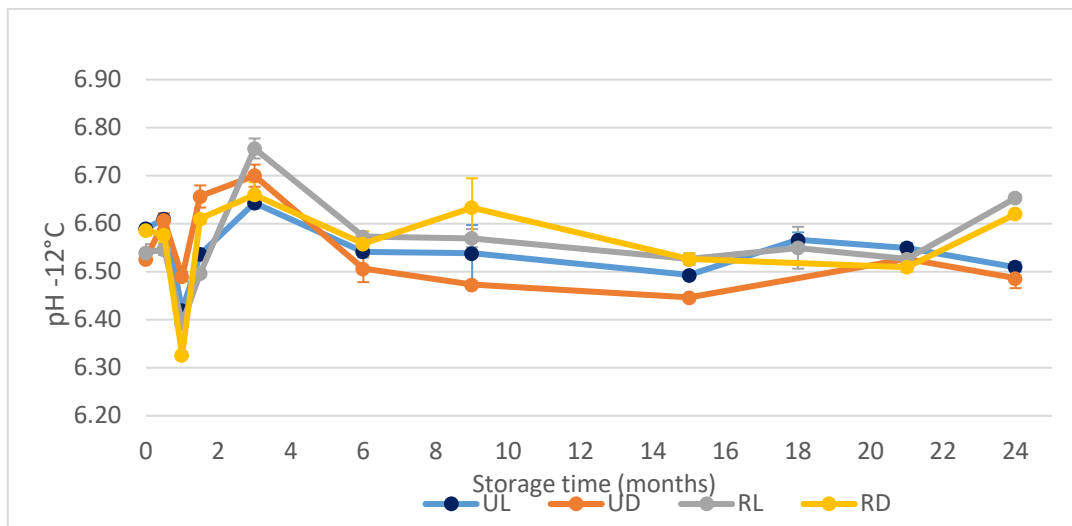


Figure 29. pH value in UL: unglazed light muscle; RL: glazed light muscle; UD: unglazed dark muscle and RD: glazed dark muscle in redfish, stored at -12°C.

The pH of the light muscle in redfish stored at -25°C was between 6.5 -6.8, and also in the dark muscle (Figure 30). Thus in most cases no significant difference was observed in muscle pH neither due to storing temperature, muscle types or treatment.

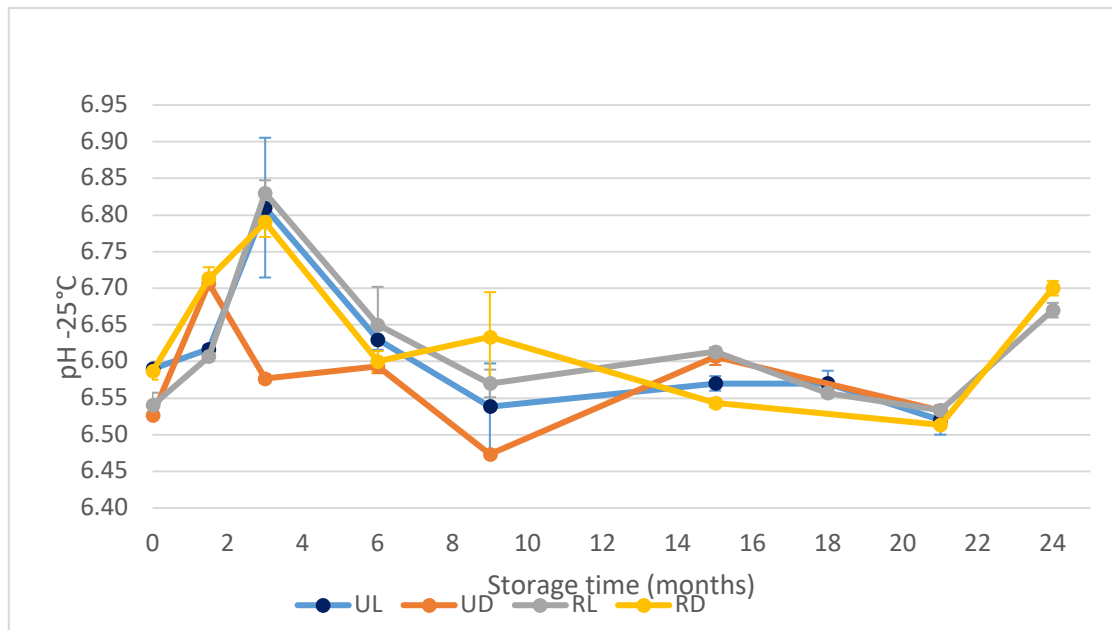


Figure 30. pH value in UL: unglazed light muscle; RL: glazed light muscle; UD: unglazed dark muscle and RD: glazed dark muscle in redfish, stored at -25°C.

Water activity (a_w)

Water activity (a_w) in the light muscle of redfish stored at -12°C was between 0.994 – 0.995, for both treatments, while the a_w in the dark muscle was between 0.994 – 0.997 for both treatments (Figure 31).

Water activity (a_w) in the light muscle of redfish stored at -25°C was between 0.992 – 0.996, for both treatments, while the a_w in the dark muscle was between 0.994 – 0.995 for both treatments (Figure 32).

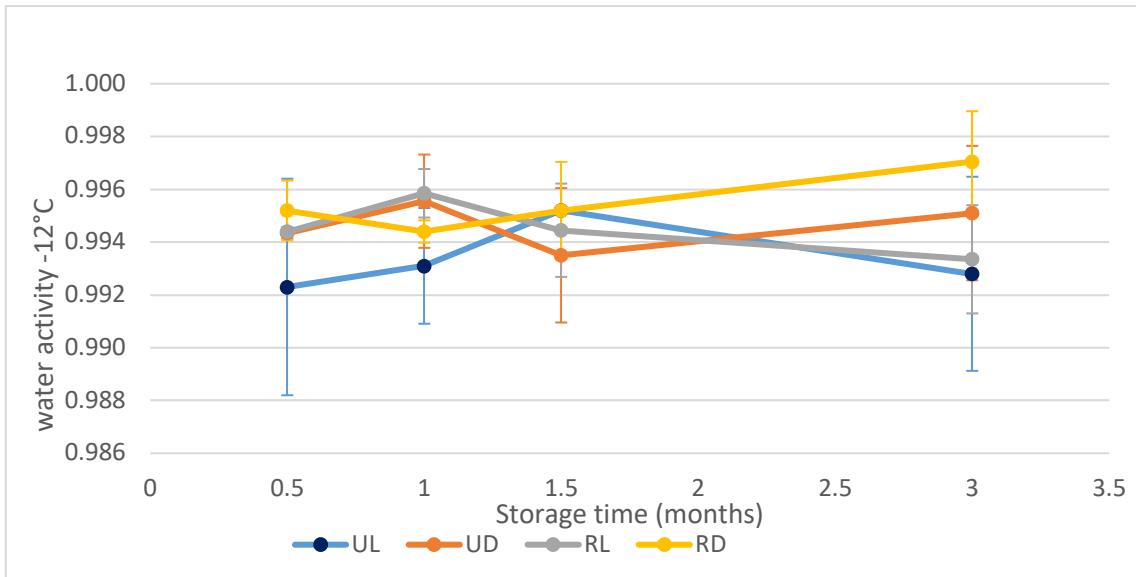


Figure 31. Water activity (a_w) in UL: unglazed light muscle; RL: glazed light muscle; UD: unglazed dark muscle and RD: glazed dark muscle in redfish, stored at -12°C.

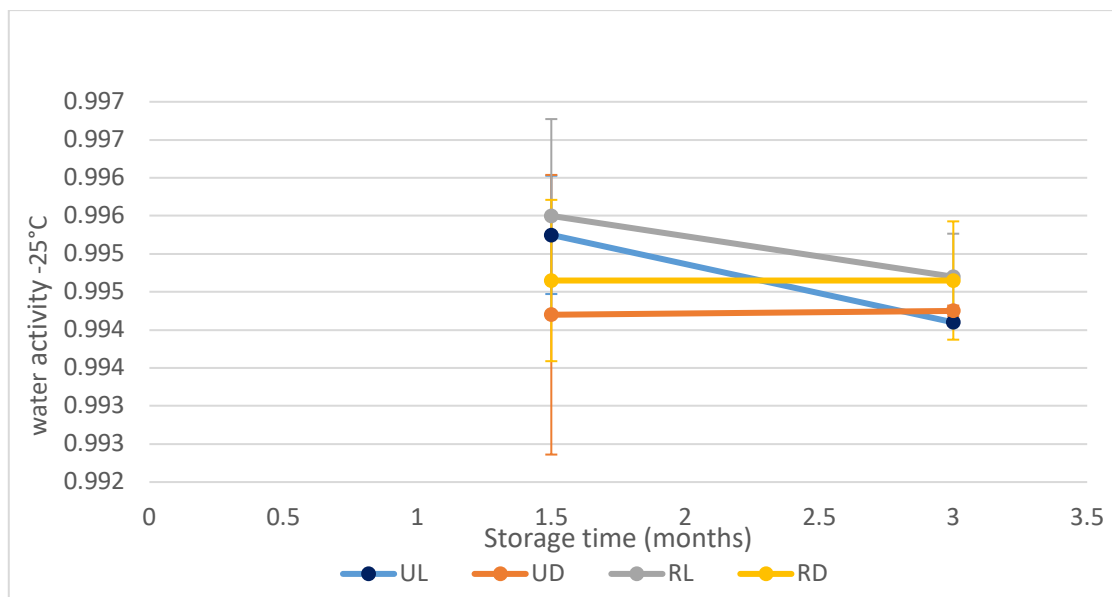


Figure 32. Water activity (a_w) in UL: unglazed light muscle; RL: glazed light muscle; UD: unglazed dark muscle and RD: glazed dark muscle in redfish, stored at -25°C.

Drip loss

The drip loss was only measured in glazed and unglazed light muscle. The drip loss was slightly higher in the light muscle in redfish stored at -12°C, compared with light muscle stored at -

25°C (Figure 33 and 34). None significant difference was observed between glazed and unglazed muscles.

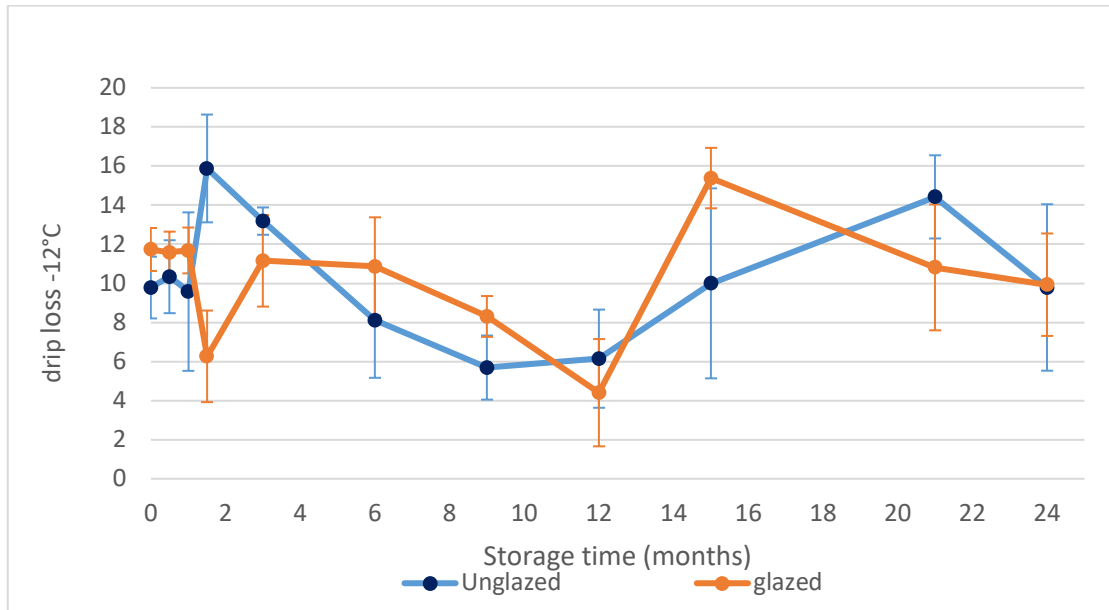


Figure 33. Drip loss (%) in UL: unglazed light muscle; RL: glazed light muscle in redfish, stored at -12°C.

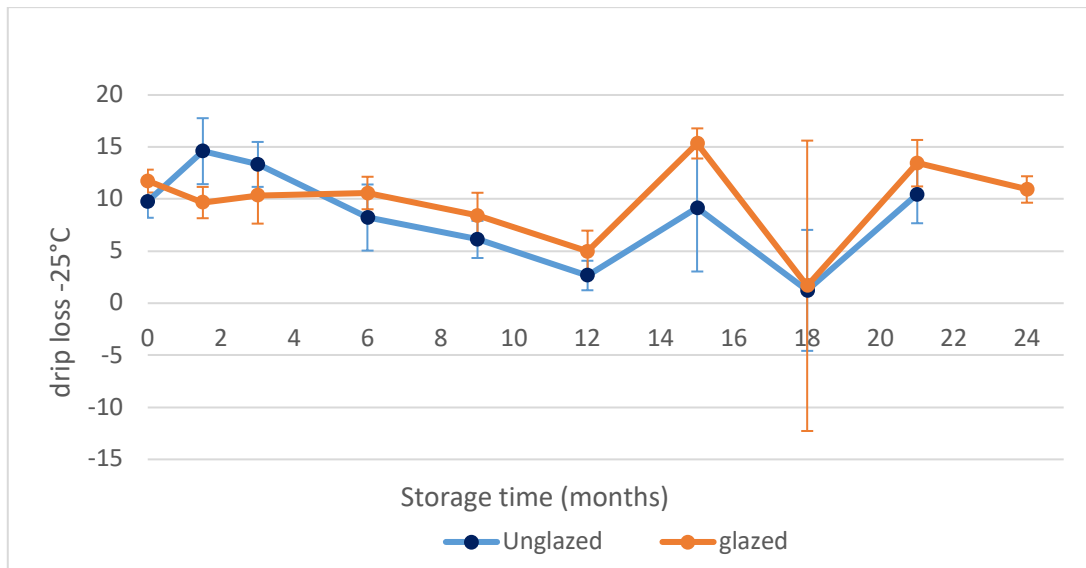


Figure 34. Drip loss (%) in UL: unglazed light muscle; RL: glazed light muscle in redfish, stored at -25°C.

Ice percentage

None significant difference in ice percentage was observed in glazed and unglazed light muscles in redfish stored at -12°C and -25°C (Figure 35 and 36). Ice percentage fluctuated slightly in the light muscle for both storing temperatures.

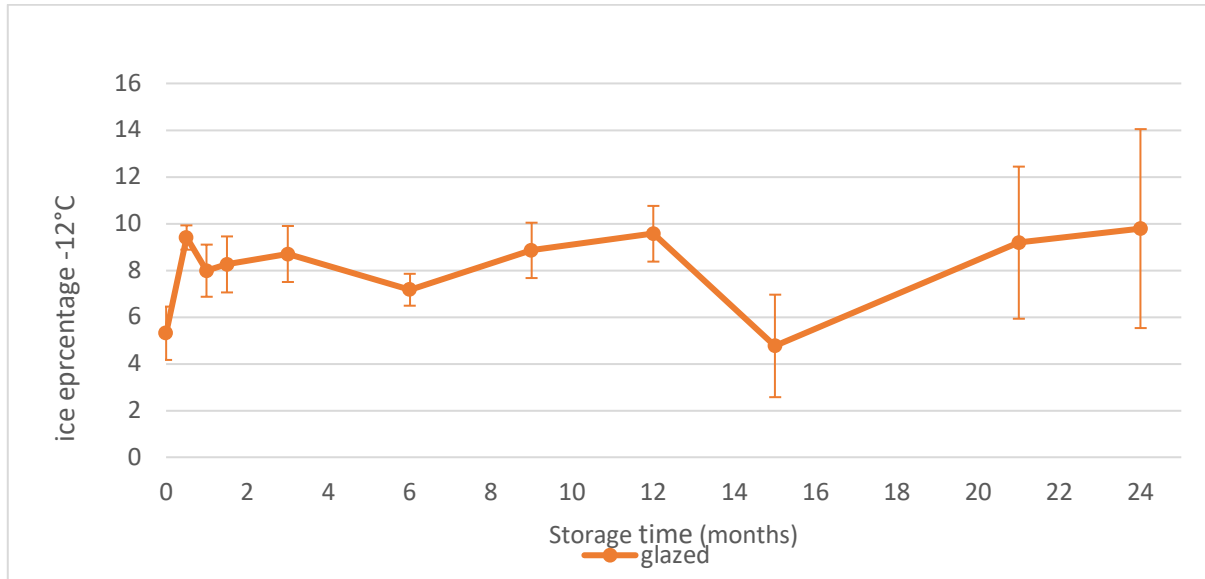


Figure 35. Ice percentage (%) in glazed light muscle in redfish, stored at -12°C .

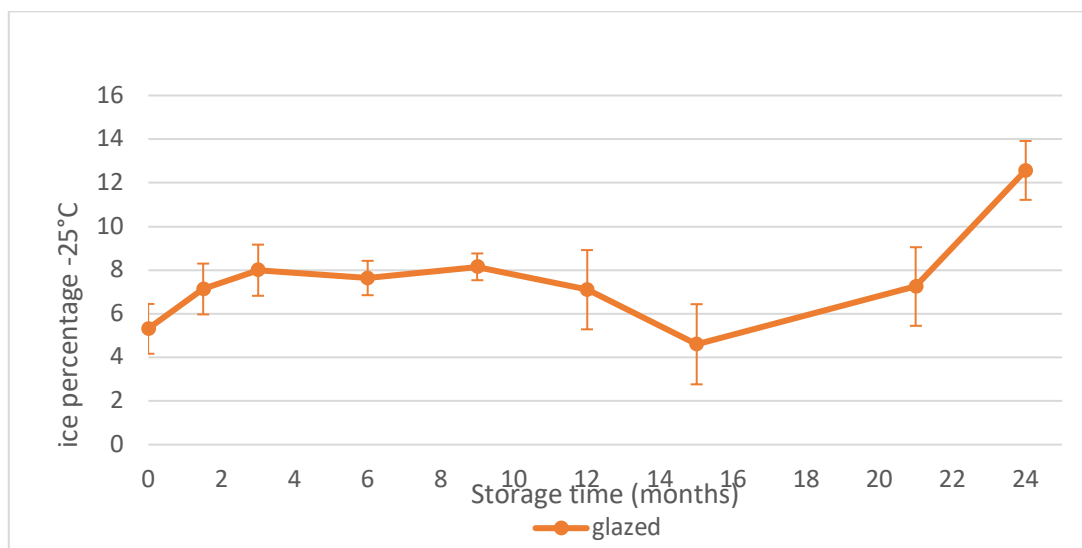


Figure 36. Ice percentage (%) in glazed light muscle in redfish, stored at -25°C .

Protein content

No significant difference was observed in protein content between glazed and unglazed light muscle in redfish stored at -12°C for most of the storage time (Figure 37). Unglazed light muscle was slightly higher from month 9 to month 18 of storage. The protein content in glazed and unglazed light muscle was rather stable during the storage period.

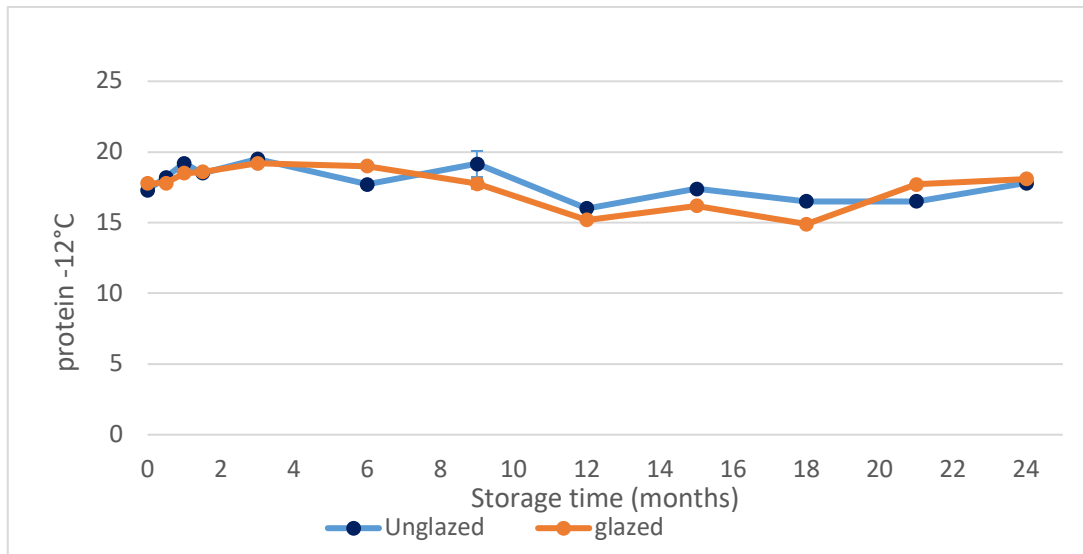


Figure 37. Protein content (%) in UL: unglazed light muscle; RL: glazed light muscle in redfish, stored at -12°C.

No significant difference was observed between glazed and unglazed light muscle in redfish stored at -25°C for most of the storage time (Figure 38). Unglazed light muscle was slightly higher from month 6 to month 12 of storage. The protein content in glazed and unglazed light muscle was rather stable during the storage period.

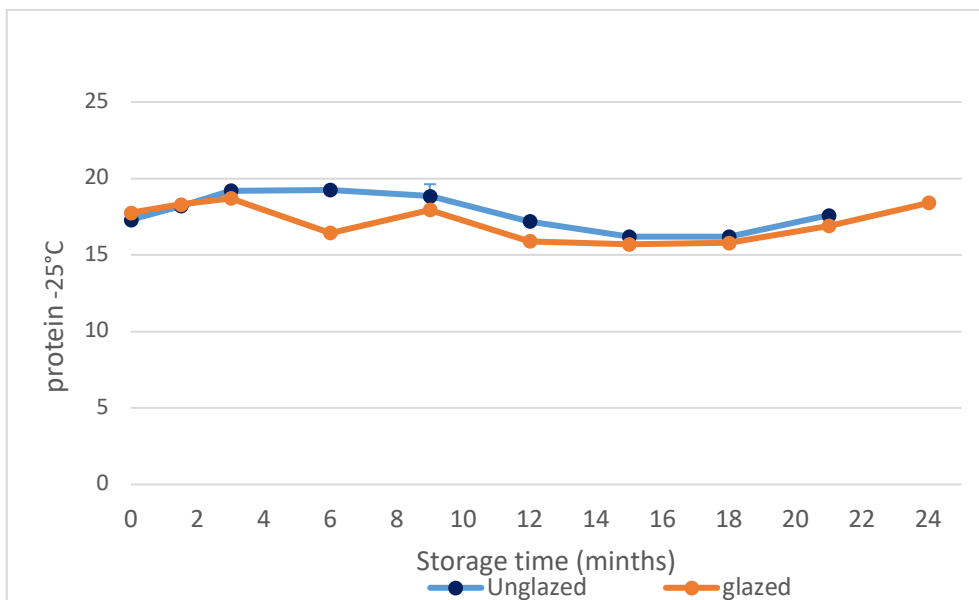


Figure 38. Protein content (%) in UL: unglazed light muscle; RL: glazed light muscle in redfish, stored at -25°C.

Total volatile basic nitrogen (TVB-N)

Total volatile basic nitrogen (TVB-N) is one of the indicators of muscle freshness. TVB-N content in glazed and unglazed light muscle fluctuated slightly during the storage of redfish stored at -12°C (Figure 39). The TVB-N content ranged between 5.8 and 9.5 mg N/100 g muscle at average. These values are within the limit of acceptance, as the critical limits of TVB-N range from 25.0 to 35.0 mg N/100 g muscle (Venugopal, 2006). After 16 and 18 months of storage the TVB-N content increased in glazed and unglazed light muscle.

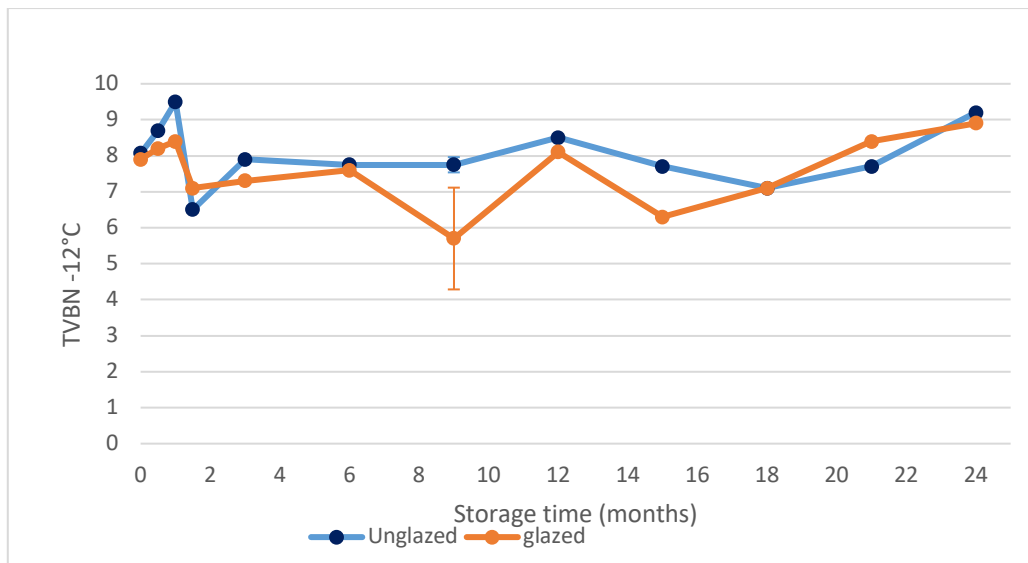


Figure 39. TVB-N content (%) in UL: unglazed light muscle; RL: glazed light muscle in redfish, stored at -12°C.

TVB-N content in glazed and unglazed light muscle was stable during the storage of redfish stored at -25°C (Figure 40). The TVB-N content ranged between 6.8 and 10.8 mg N/100 g muscle at average. After 15 months of storage the TVB-N content increased in glazed and unglazed light muscle.

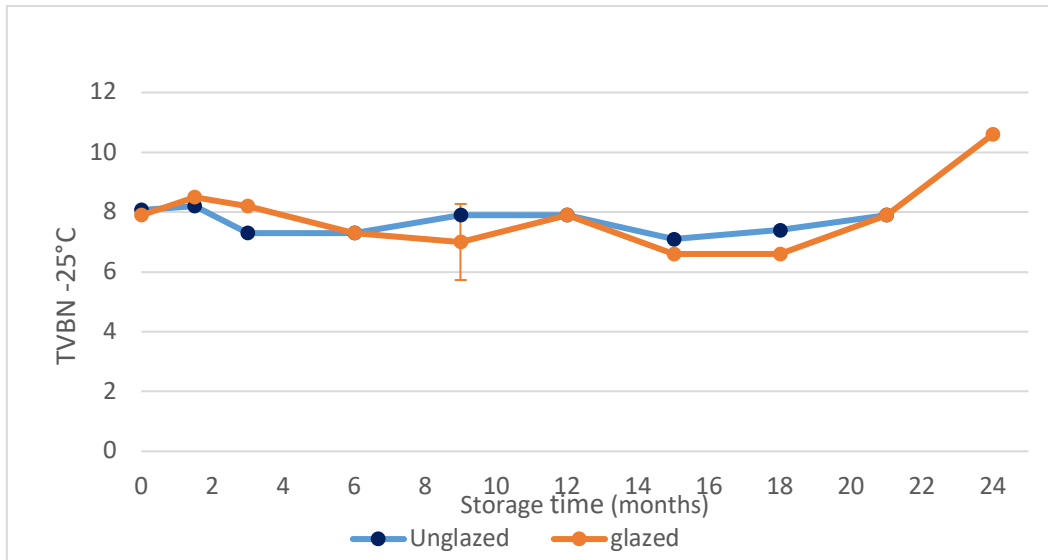


Figure 40. TVB-N content (%) in UL: unglazed light muscle; RL: glazed light muscle in redfish, stored at -25°C.

Water holding capacity (WHC)

The ability of muscle to retain water (WHC) is an essential quality parameter. The WHC in glazed and unglazed light muscle decreased during storing for 12 months at -12°C (Figure 41). Then WHC remained stable to the end of the storing period. There were no significant difference between glazed and unglazed light muscle. The decrease in WHC on the early stage of storage can likely be explained by the protein denaturation in the redfish muscle, occurring during frozen storage, which led to a loss of functional properties of the proteins. Moreover, enzymatic activities (lipase, phospholipase and protease) in the light muscle are also thought to contribute to the decrease in WHC (Xiong, 1997).

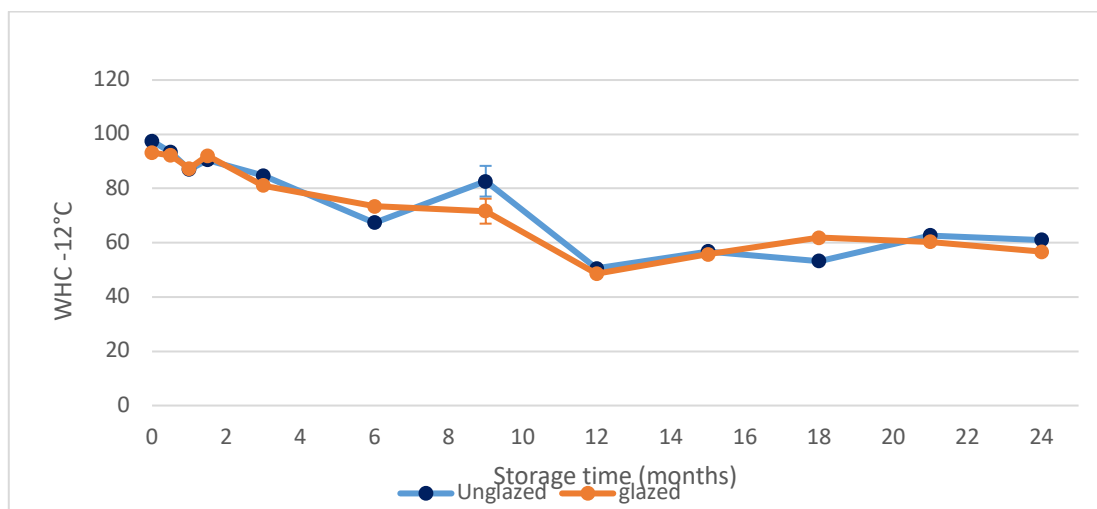


Figure 41. Water holding capacity (%) in UL: unglazed light muscle; RL: glazed light muscle in redfish, stored at -12°C.

The WHC in glazed and unglazed light muscle decreased during storing for 12 to 15 months at storing temperature of -25°C (Figure 42). The WHC remained stable in the unglazed light muscle to the end of the storage, while WHC increased in glazed light muscle to the end of the storage.

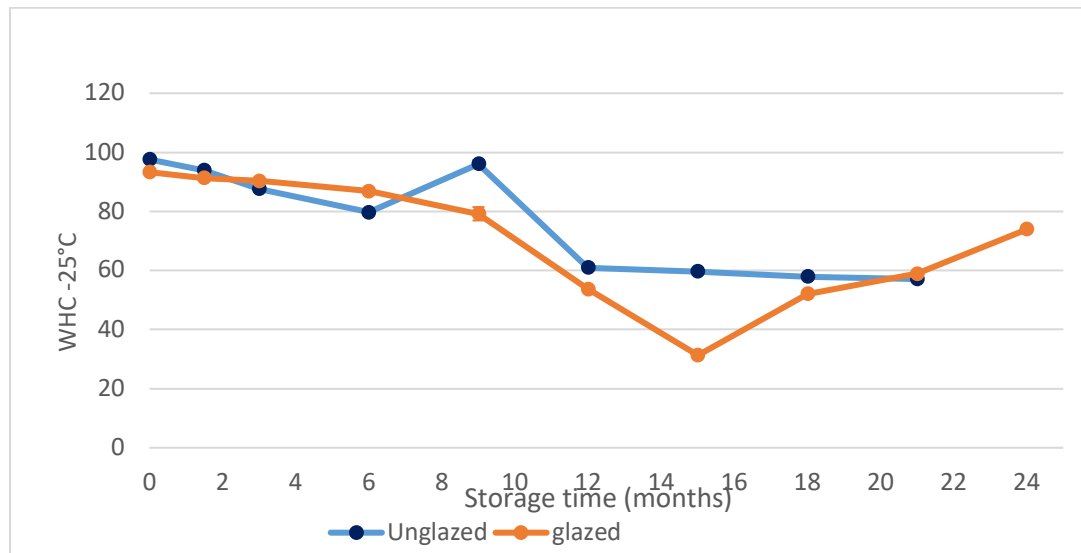


Figure 42. Water holding capacity (%) in UL: unglazed light muscle; RL: glazed light muscle in redfish, stored at -25°C.

Raw materials, processing and sampling (June and November 2015)

Thawing loss, WHC and CY

Thawing of fillets from fish caught in June was fairly stable throughout the storage period, while an increase in thawing loss was observed for the fish caught in November (Figure 43a). The increase in thawing loss was considered to be mainly due to protein denaturation (Xiong, 1997) and cell rupture caused by ice crystal formation. Processing of raw material of lower freshness (on day 9) led to a slightly higher thawing loss compared to fillets processed and frozen 4 days postcatch for both seasons. The higher thawing loss observed in the fish processed on day 9 can also be assumed to be mainly due to higher protein denaturation that occurs during ice storage, as freshness affects the biochemical properties of fish muscle (Kim & Park, 2007).

The ability of muscle to retain water (WHC) is an essential quality parameter. The WHC of the light muscle was stable during the first 4 months, followed by a decrease until month 8 and month 12 of storage for fish caught in both seasons (Figure 43b). The WHC then remained stable to the end of the study. The decrease in WHC on the early stages of storage can likely

be explained by the protein denaturation in the fish muscle occurring during frozen storage, which led to loss of functional properties of the proteins. Moreover, enzymatic activities (lipase, phospholipase and protease) in the light muscle are also thought to contribute to the decrease in WHC (Xiong, 1997).

The CY is related to the liquid lost during cooking and comes from constitutive water as a result of protein denaturation and from the fat which melts during heating. CY increased during the first 8 months of storage for all treatments (Figure 43c). A significant negative correlation ($r = -0.64$) was obtained between the WHC and the CY of the fillets. No significant differences were observed in the CY neither due to the raw material freshness nor the season of capture.

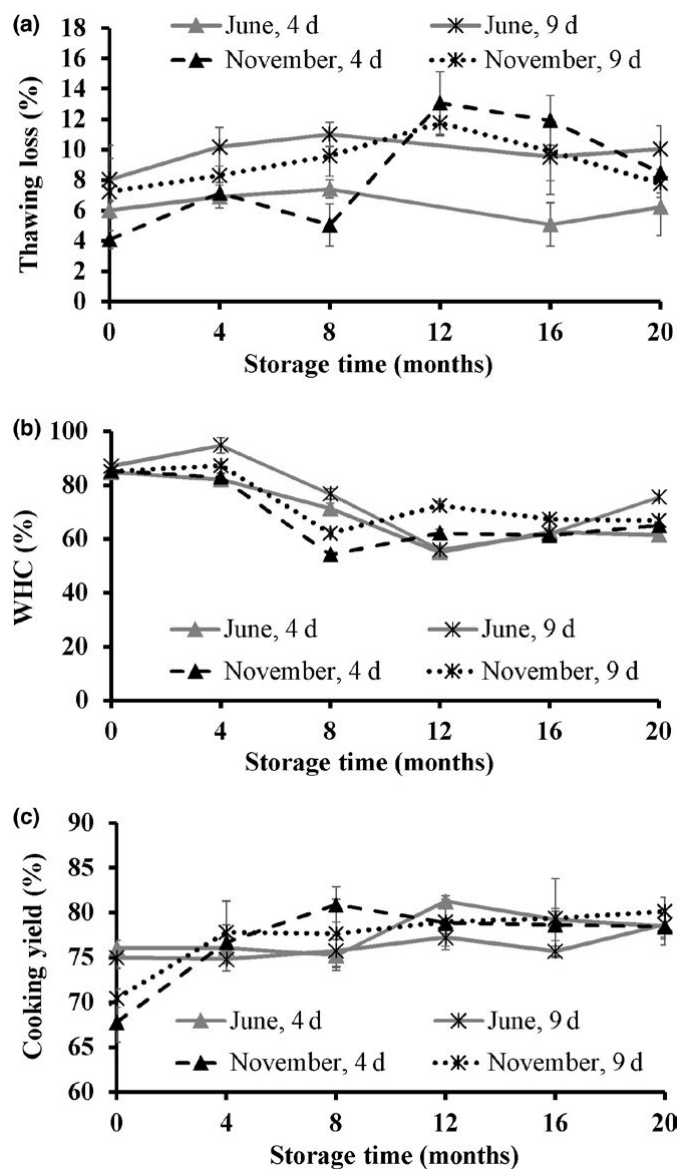


Figure 43. Thawing loss (%) (a), water holding capacity (WHC; %) (b), and cooking yield (%) (c) of golden redfish fillets as affected by the seasonal variation (June and November), and raw material freshness (processed 4 and 9 days postcatch) through frozen storage at -25°C ($n=3$, mean \pm standard deviation).

Color

Lighness values of the golden redfish fillets were stable during the frozen storage for all treatments. Furthermore, no significant differences were observed in the L values neither due to freshness nor catching season (Figure 44a). The a -value of the fillets processed 4 days postcatch in June fluctuated slightly, but in general, the a -value of the fillets was stable for both seasons (Figure 44b). In general, the b -values increased during frozen storage, indicating a more yellow appearance of the fillets with storage. Slightly higher b -values were observed in the fish processed 9 days postcatch than in the fish processed 4 days postcatch (Figure 44c), in agreement with the lower freshness of the fish processed 9 days postcatch.

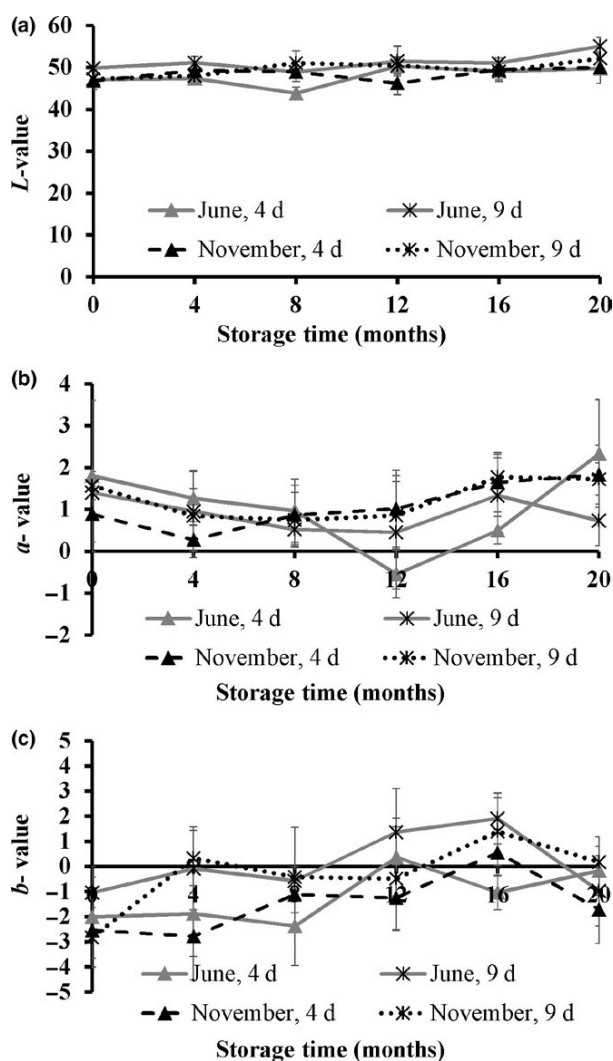


Figure 44. Color L-value (a), a-value (b) and b-value (c) of redfish fillets as affected by the seasonal variation (June and November) and raw material freshness (processed 4 and 9 days postcatch) through frozen storage at -25°C (n = 3, mean ± standard deviation).

Weight proportions, proximate composition and PL content

The light muscle constituted $88.1 \pm 2.6\%$ of the fillets, independent of the season. The water content in the light and dark muscle of the fish caught in June (processed 4 days postcatch) was $80.2 \pm 0.3\%$ and $75.4 \pm 0.3\%$, respectively. However, significantly higher water content was observed in the fish caught in November, as represented by a water content of $81.4 \pm 0.2\%$ and 77.7 ± 0.4 , in the light and dark muscle, respectively. The TL content of the light muscle was significantly lower than in the dark muscle of fish caught in both seasons. The protein content of the light muscle was $18.2 \pm 0.1\%$ in June and $18.9 \pm 0.1\%$ in November. According to Gíslason and Ástþórsson (1995), the springbloom of the phytoplankton and total zooplankton in the southwest of Iceland are low in the winter and reach a maximum during the summer (May-June). In this study, golden redfish were caught at these places, which could

explain that even though the redfish were caught in the late spawning season (June), the protein and TL content of the muscle did not differ from fish in the mating season (November) due to heavy feeding.

The water content in the light muscle was both higher and more stable during frozen storage compared to the dark muscle of both seasons and the raw material freshness upon processing and freezing (4 or 9 days postcatch) (Figure 45a,b). The water content in the dark muscle of the redfish caught in June was initially stable but then tended to decrease after 12 months of frozen storage. The redfish caught in November, the water content in the dark muscle fluctuated throughout the storage time, and exhibited a lower water content than the fish caught in June. The water content and TL content were inversely correlated ($r = -0.96$) throughout storage and constituted together around 80.0% - 82.0%, which is in agreement with the findings of Karlsdóttir *et al.* (2014) on the composition of saithe and hoki. The TL content of the light muscle was stable, but fluctuated heavily in the dark muscle during frozen storage.

Higher TL values were generally observed in redfish caught in November compared to June (Figure 45c,d). The protein content was stable during the frozen storage of both raw materials freshness states upon processing and season of both catch.

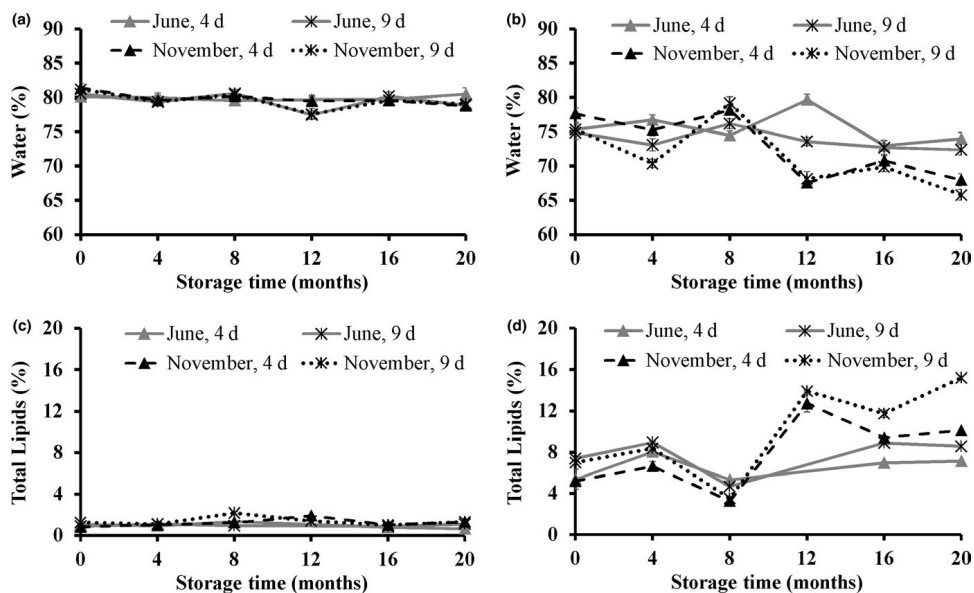


Figure 45. water content in the light(a) and the dark (b) muscle, and a total lipids content in the light (c) and the dark (d) muscle of redfish, as affected by the seasonal variation, and raw material freshness (processed 4 and 9 days postcatch) through frozen storage at -25°C ($n = 3$, mean \pm standard deviation).

The PL content was significantly higher in the light muscle compared with the dark muscle (Figur 46a,b) in agreement with previous studies of saithe and hoki (Karlsdóttir *et al.*, 2014) and herring (Dang *et al.*, 2017). No significant differences were observed in PL content of the light muscle due to raw material freshness. Higher PL content was generally observed in the light muscle of fish caught in June than in November. The PL content in the light muscle of fish caught in June increase during the first 4 months of storage, which may be attributed during storage (Kolowska *et al.*, 2003). After 4 months storage, the PL content decreased for the remaining duration of storage., the PL content of the light muscle of fish caught in November decreasec throughout the frozen storage. This decrease in PL content in both seasons after long term frozen storage was expected due to the hydrolytic activites by phospholipases (Sista, *et al.*, 1997). No signifant changes were observed in PL content in the dark muscle, independent of season of catch.

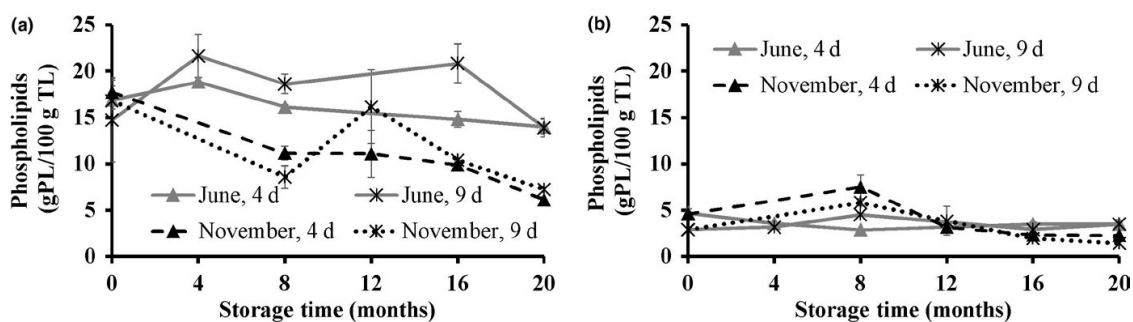


Figure 46. Phospholipids content (gPL/100g TL) in the light (a) and the dark (b) muscle of redfish, as affected by the seasonal variation, and raw material freshness (processed 4 and 9 days postcatch) through frozen storage at -25°C (n= 3, mean ± standard deviation).

Fatty acid profile

Polyunsaturated fatty acids (PUFA) were dominant in the fatty acid profile in the redfish light muscle of the raw material of both seasons, followed by monosaturated fatty acids (MUFA) and finally saturated fatty acids (SFA) (Table 3). In the dark muscle, the amount of MUFA were the highest, followed by PUFA, and lastly SFA. These findings were similar to the previous observation of the fatty acid composition of hoki by Karlsdóttir *et al.* (2014).

The major SFA in both muscle types and seasons was palmitic acid (C16:0), while erucic acid (C22:1n-9) and oleic acid (C18:1n-9) were the predominant fatty acids amongst the MUFAs. Significantly higher amounts of oleic acid were observed in both muscle types of fish captured

Table 3. Fatty acid profile (g fatty acids/100 g of golden redfish light and dark muscle (analyzed 4 days postcatch) caught in Icelandic waters in June and November, 2015 (n= 3, mean \pm standard deviation).

Fatty acids	Light muscle		Dark muscle	
	June	November	June	November
C14:0	3.4 \pm 0.4 ^{ax}	2.6 \pm 0.4 ^{bx}	5.0 \pm 0.2 ^{zy}	4.3 \pm 0.3 ^{by}
C15:0	0.3 \pm 0.0 ^a	0.2 \pm 0.1 ^{ax}	ND	0.3 \pm 0.0 ^x
C16:0	15.6 \pm 0.8 ^{ax}	15.8 \pm 0.3 ^{ax}	12.4 \pm 0.1 ^{zy}	12.6 \pm 0.2 ^{zy}
C17:0	0.2 \pm 0.0 ^{ax}	0.1 \pm 0.1 ^{ax}	0.2 \pm 0.0 ^{ax}	0.1 \pm 0.0 ^{ax}
C18:0	3.0 \pm 0.1 ^{ax}	3.2 \pm 0.0 ^{ax}	2.6 \pm 0.0 ^{zy}	2.7 \pm 0.1 ^{zy}
Σ SFA	22.5 \pm 1.0 ^{ax}	21.9 \pm 0.1 ^{ax}	20.4 \pm 0.9 ^{zy}	20.1 \pm 0.0 ^{zy}
C16:1n9	3.7 \pm 0.4 ^{ax}	3.9 \pm 0.1 ^{ax}	4.9 \pm 0.7 ^{zy}	5.9 \pm 0.4 ^{by}
C18:1n5	1.3 \pm 0.6 ^x	ND	2.4 \pm 0.8 ^y	ND
C18:1n7	4.2 \pm 1.2 ^{ax}	2.7 \pm 0.0 ^{bx}	5.7 \pm 3.6 ^{zy}	3.3 \pm 0.2 ^{by}
C18:1n9	7.0 \pm 1.3 ^{ax}	9.7 \pm 0.2 ^{bx}	8.1 \pm 4.6 ^{zy}	13.1 \pm 1.4 ^{by}
C20:1n7	4.7 \pm 0.5 ^{ax}	7.8 \pm 0.2 ^{bx}	9.0 \pm 2.1 ^{zy}	11.6 \pm 0.2 ^{by}
C20:1n9	0.9 \pm 0.7 ^{ax}	0.3 \pm 0.1 ^{ax}	2.9 \pm 1.8 ^{zy}	0.4 \pm 0.2 ^{bx}
C20:1n11	1.4 \pm 0.7	ND	ND	0.8 \pm 0.1
C22:1n9	7.9 \pm 2.2 ^{ax}	7.7 \pm 0.2 ^{bx}	13.0 \pm 1.1 ^{zy}	11.9 \pm 0.7 ^{by}
C22:1n11	2.1 \pm 0.9 ^{ax}	1.1 \pm 0.0 ^{bx}	2.6 \pm 1.1 ^{zy}	1.7 \pm 0.2 ^{by}
C24:1	0.6 \pm 0.1 ^{ax}	0.2 \pm 0.0 ^{bx}	2.0 \pm 1.2 ^{zy}	0.3 \pm 0.0 ^{bx}
Σ MUFA	32.5 \pm 3.2 ^{ax}	33.5 \pm 0.2 ^{ax}	49.5 \pm 0.0 ^{zy}	48.4 \pm 0.6 ^{by}
C16:3n3	0.6 \pm 0.0 ^x	ND	0.5 \pm 0.3 ^x	ND
C18:3n3	1.3 \pm 0.1 ^{ax}	0.4 \pm 0.0 ^{bx}	1.7 \pm 0.6 ^{zy}	0.5 \pm 0.0 ^{by}
C18:4n3	1.0 \pm 0.1 ^{ax}	0.7 \pm 0.3 ^{ax}	1.3 \pm 0.0 ^{zy}	1.2 \pm 0.4 ^{by}
C20:4 n3	ND	0.4 \pm 0.0 ^x	ND	0.5 \pm 0.0 ^x
C22:5 n3	ND	0.4 \pm 0.0 ^x	ND	0.3 \pm 0.3 ^x
C20:5n3 (EPA)	7.9 \pm 0.3 ^{ax}	8.3 \pm 0.2 ^{bx}	6.5 \pm 1.6 ^{zy}	7.3 \pm 0.0 ^{y^a}
C22:6n3 (DHA)	23.5 \pm 1.1 ^{ax}	26.3 \pm 0.6 ^{bx}	11.5 \pm 1.3 ^{zy}	14.5 \pm 0.0 ^{by}
Σ n-3	34.2 \pm 0.6 ^{ax}	36.5 \pm 0.7 ^{bx}	21.4 \pm 0.0 ^{zy}	24.3 \pm 0.4 ^{by}
C18:2n6	1.7 \pm 0.2 ^{ax}	1.6 \pm 0.1 ^{ax}	1.5 \pm 0.2 ^{ax}	1.5 \pm 0.0 ^{bx}
C20:4n-6	ND	1.2 \pm 0.0 ^x	ND	0.7 \pm 0.1 ^y
C22:5n6	1.1 \pm 0.2 ^{ax}	0.9 \pm 0.0 ^{ax}	1.4 \pm 0.9 ^{ax}	0.8 \pm 0.1 ^{bx}
Σ n-6	2.7 \pm 0.4 ^{ax}	3.7 \pm 0.1 ^{bx}	2.9 \pm 1.1 ^{ax}	3.1 \pm 0.1 ^{by}
C16:2n4	0.4 \pm 0.1 ^{ax}	0.2 \pm 0.0 ^{bx}	0.8 \pm 0.0 ^{zy}	0.2 \pm 0.0 ^{bx}
C20:4	0.5 \pm 0.1 ^x	ND	0.6 \pm 0.0 ^x	ND
Σ PUFA	37.9 \pm 0.1 ^{ax}	40.4 \pm 0.7 ^{bx}	25.7 \pm 1.0 ^{zy}	27.6 \pm 0.5 ^{by}
n-3/n-6	12.7 \pm 2.2 ^{ax}	9.8 \pm 0.3 ^{bx}	7.9 \pm 2.9 ^{zy}	8.0 \pm 0.2 ^{zy}

a,b: different superscript letters in each row indicate a significant differences within a variable between seasons (June vs November) in the same muscle types (light or dark muscle).

x,y: different superscript letters in each row indicate a significant differences within a variable between muscle types (light vs dark) within the same season of catch.

ND: not detected.

in November compared to fish caught in June. Amongst the PUFAs, docosahexaenoic acid (DHA, C22:6n-3) predominated the profile, followed by eicosapentaenoic acid (EPA, C20:5n-3). EPA og DHA levels were higher in the light muscle than in the dark muscle of the redfish at both seasons. Furthermore, a higher n-3/n-6 was obtained in the light muscle compared to the dark muscle of fish caught during both seasons. These results indicated that the light

muscle can be considered more valuable than the dark muscle, as the *n-3/n-6* ratio is a good index for comparing the relative nutritional value of fish oils (Pigott & Tucker, 1990), but a higher *n-3/n-6* ratio in the human dietary has been shown to have positive traits toward preventing coronary heart disease and in reducing cancer risk (Kinsella *et al.*, 1990).

Generally, no significant differences were observed due to season of catch in the amount of SFAs and MUFAs of neither muscle types. The amount of PUFAs in the fish caught in November was significantly higher than in the fish caught in June, and especially in the amounts of EPA and DHA. These differences are thought to be mainly influenced by the season, food availability, age and size of the fish, as well as their maturation status (Aidos *et al.*, 2002). The higher amount of PUFA in the fish caught in November might contribute to the higher instability of the lipid properties during frozen storage of the fish caught in November, compared to the fish caught in June. These findings were in line with the observation of Dewitt (1963) who reported that the unsaturation of cod liver oil increased steadily from summer to autumn and reached a maximum in the winter.

Muscle pH and volatile basic nitrogen

The initial muscle pH was 6.6 ± 0.2 for both the light and dark muscle of fish caught in June, whereas a pH of 6.7 ± 0.1 in both muscle types in the fish caught in November. Thus, no significant difference was observed in muscle pH neither due to muscle types, raw material freshness (processed 4 and 9 days postcatch), nor season. Furthermore, the frozen storage did not result in any significant pH changes, ranging from 6.5 to 6.7 for both muscle types of fish caught in June, and from 6.6 to 6.8 for fish caught in November.

Total volatile basic nitrogen is one of the indicators of muscle freshness. The TVB-N fluctuated during frozen storage and ranged between 11.0 and 14.0 mg N/100 g in the light muscle, independent of season and raw material freshness. These values are within the limit of acceptance, as the critical limits of TVB-N range from 25.0 to 35.0 mgN/100 g muscle (Venugopal, 2006). These results indicated that the light muscle of the redfish fillets stored at -25°C was stable and of acceptable freshness in terms of TVB-N, during the 20 months of storage.

Frozen storage inhibited microbial activity, which corresponds basically to both pH and TVB-N values. This could explain the low values of both pH and TVB-N obtained in the present study.

Lipid oxidation and hydrolysis

Lipid oxidation was assessed by the analysis of primary (PV) and secondary (TBARS) lipid oxidation products. Significant increase in both PV and TBARS content of both muscle types were observed in all treatments during frozen storage. However, the formation rate of oxidation products in the fish fillets depended strongly on the muscle types. The dark muscle was more progressive toward lipid oxidation compared to the light muscle. This was expected due to the higher lipid content in the dark muscle compared to the light muscle (Hultin, 1994). A similar trend has been observed earlier for both lean fish (Karlsdóttir *et al.*, 2014) and fatty fish species (Dang *et al.*, 2017; Underland *et al.*, 1998).

In the light muscle, the formation of PV increased only after 4 months, reaching a small peak after 8 months of frozen storage (Figure 47a). In the dark muscle, the PV content increased sharply already from the beginning of storage and reached a peak at 4 and 8 months for the fish caught in June and in November, respectively (Figure 47b). The earlier oxidation production in the dark muscle of fish caught in November is thought due to its higher PUFA content compared to fish caught in June.

The peaks in PV were followed by decomposition of hydroperoxides to secondary oxidation products until month 12 and 16 of frozen storage for the June and November samples, respectively. This was coupled to an increase in TBARS content observed at the same storage points. The TBARS content of the light muscle increased slowly during the storage time (Figure 47c). However, the TBARS content in the dark muscle of fish caught in June fluctuated, but with an overall increasing trend during storage for fish caught at both seasons (Figure 47d). Pearson's correlation showed a significant positive correlation ($r = 0.53$) between TBARS content and yellowness. The increase in b -value is thus believed to be related to oxidation of the muscle lipids during frozen storage (Erickson, 1977), as indicated by an increasing yellow/brown nuance. A significant negative correlation ($r = -0.71$) was obtained between the PV content and WHC of the light muscle. This indicates that lipid oxidation may influence WHC of the muscle, either directly or indirectly, through oxidative alteration of the muscle proteins.

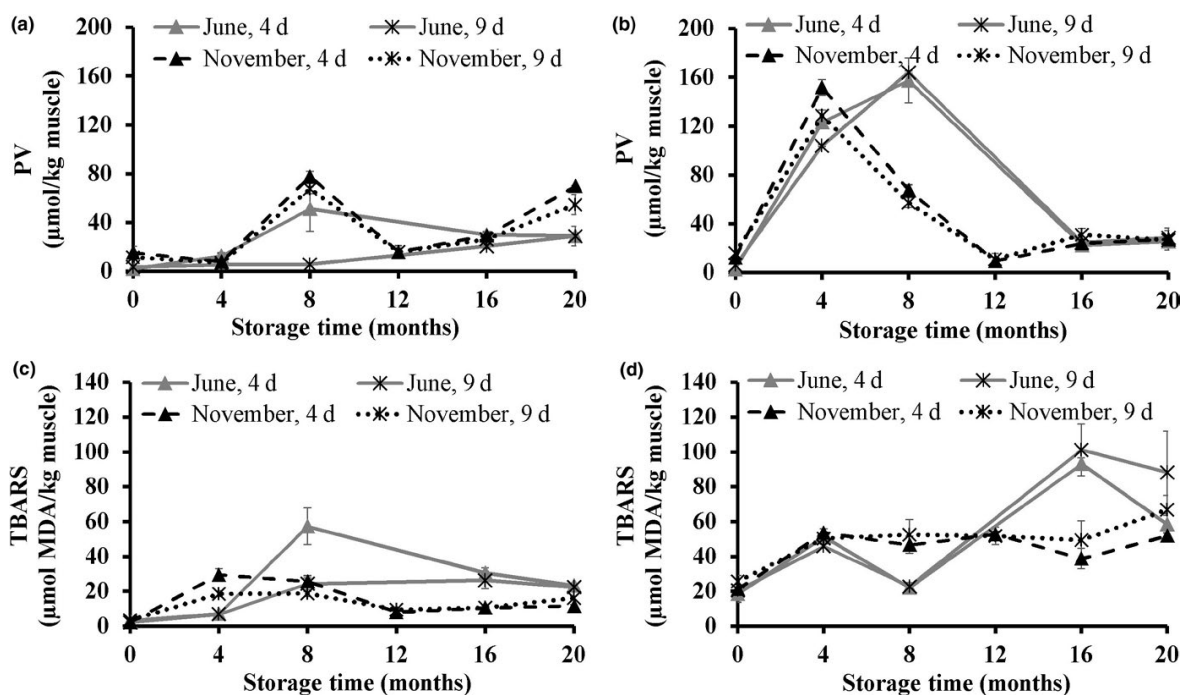


Figure 47. Peroxide value (PV; $\mu\text{mol}/\text{kg}$ muscle) in the light (a) and the dark (b) muscle, and thiobarbituric acid reactive substances (TBARS; μmol MDA/kg muscle) in the light (c) and the dark (d) muscle of golden redfish, as affected by the seasonal variation (June and November), and a raw material freshness (processed 4 and 9 days postcatch) through frozen storage at -25°C ($n=3$, mean \pm atandard deviation).

Lipid hydrolysis may occur postmortem in fish leading to an increase in FFA content, due to increased lipase and phospholipase activities (Pacheco-Aguilar *et al.*, 2000). Higher hydrolytic activity as assessed by FFA content was observed in the light muscle compared to the dark muscle at both seasons (Figure 48). Accumulation of FFA was observed in the light muscle during storage, while the FFA formation was rather stable in the dark muscle. This can be explained by a higher PUFA content in the light muscle, making it more susceptible to lipid hydrolysis compared to the dark muscle (Polvi *et al.*, 1991). Faster formation of FFA in the light muscle during frozen storage may be a consequence of lipid degradation due to phospholipase and lipase activity (Auborg & Medina, 1999; Pacheco-Aguilar *et al.*, 2000), which may, furthermore, explain the observed reduction of PL content in the light muscle during storage (Figure 46a).

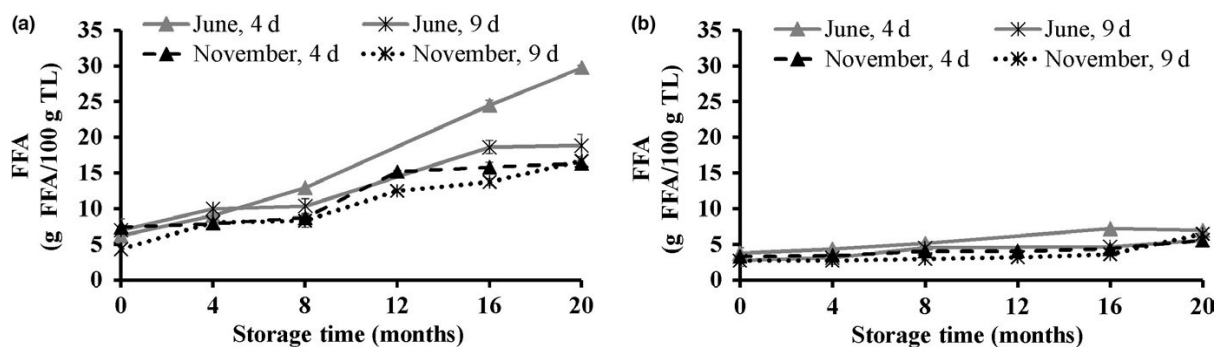


Figure 48. Free fatty acids content (FFA; g FFA/100 g TL) in the light (a) and the dark (b) muscle of golden redfish, as affected by the seasonal variation (June and November), and raw material freshness (processed 4 and 9 poistcatch) through frozen storage at -25°C ($n = 3 \pm$ standard deviation).

A significant negative correlation ($r = -0.58$) was observed between the FFA and TL content. This is in agreement with the previous studies of Sheltawy and Olley (1966) for cod muscle. On the other hand, lipid hydrolysis may speed up lipid oxidation (Kaneniwa *et al.*, 2000; Sista *et al.*, 1997), which is consistent with the increase in PV and TBARS content during frozen storage as observed in the current study. Results from the present study also show a relationship between FFA content and WHC ($r = -0.60$) in the light muscle. It can be assumed that the FFA accumulation in the light muscle affected texture deterioration of the fillet by interacting with the muscle proteins (Mackie, 1993), which led to a decrease in WHC of the light muscle during frozen storage. The formation of FFA was, however, neither affected by the season, nor the freshness of raw material.

Multivariate data analysis

A principal component analysis (PCA) was performed on the data to obtain an overview of the effects of the storage duration, raw material freshness, and season on the analyzed variables. The scores and correlation loadings from the first and second principal components (PCs) are shown in Figure 49. The PC1, representing 39% of the variance between the golden redfish samples, mostly indicated the differences in the chemical composition and lipid degradation of the light versus dark muscle types. The light muscle had a higher water content than the dark, While the dark muscle was characterized by a higher lipid content. PC2, representing 27% of the total variation, described the effects of storage duration on physical properties and lipid degradation. Thawing loss, CY, lipid hydrolysis, and oxidation increased with prolonged frozen storage, while the WHC decreased. As seen from the variable distribution on the loadings graph, the degradation of the dark muscle was dominated by lipid hydrolysis, while

the degradation of the dark muscle was dominated by lipid oxidation, in agreement with the non-variate analysis.

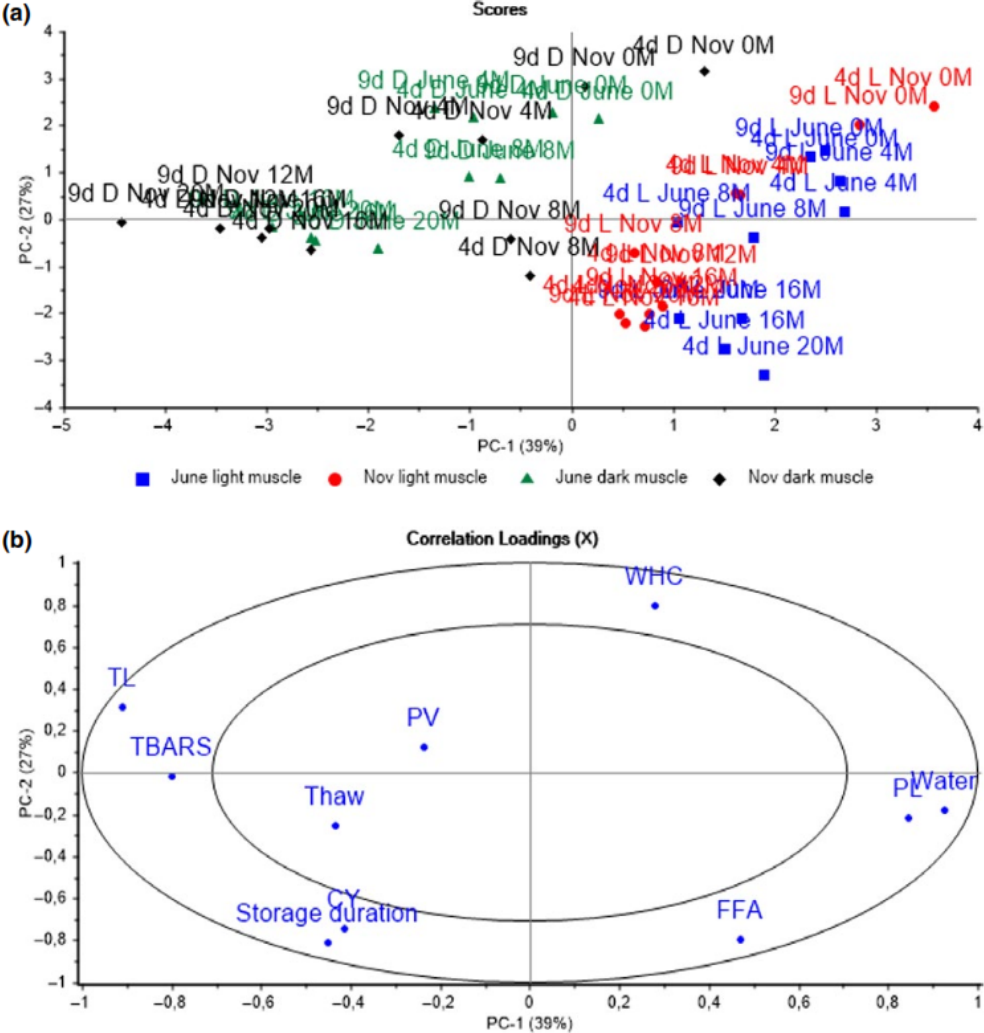


Figure 49. Scores (a) and correlation (b) from the principal component analysis (PCA) of light and dark muscles of golden redfish fillets. The first number (4 and) and second letter (d) indicate the processing delay in days. The letter L or D indicates the light and dark muscle. The number and the last letter (M) of the sample description indicate the frozen storage time in months. June and November indicate whether the fish were caught in June or November.

Conclusions

Storing temperatures affected many of the physical and chemical properties in redfish, caught in January. There was less fat content in the light muscle, compared to the dark muscle for both storing temperatures (-12°C vs -25°C). Storing temperatures affected the amount of free fatty acid. Storage at -25°C contributed in lower FA content. Also longer storage time contributed an increase of the FA. The TBARS also increased during the storage time. The light muscle was lower in TBARS value, then the dark muscle. Storage did not effect the cooking yield for both storage temperatures. Water content was higher in the light muscle, compared with the dark muscle. The drip loss was higher in glazed light muscle, compared with unglazed muscle. That was no surprise since the glazed muscle had extra water on the surface of the muscle. TVB-N increased slightly with storage in the light muscle. Water holding capacity decreased with storage time in the light muscle.

Season of capture affected both the nutritional value and stability of golden redfish. The light muscle of fish caught in November was richer in EPA and DHA than in the fish caught in June. The fish caught in November was also more unstable through frozen storage, due to a more unsaturated nature of the fatty acids present, indicating that special care needs to be applied during handling and treatment of golden redfish caught at this time. Fish processed and frozen 9 days postcatch had slightly higher thawing loss and yellowness values compared to fish processed on day 4, but no differences were found in other quality attributes.

The light muscle had a higher nutritional value than the dark muscle and is a good nutritional source for human consumption. However, the dark muscle was prone to lipid oxidation which may have a negative influence on the more valuable light muscle. Removing the dark muscle by deep skinning could thus improve the quality and stability of redfish fillets. However, the dark muscle may be used for fish meal, fish oil production, or other valuable products for human consumption, leading to a more sustainable utilization of golden redfish.

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