Contents lists available at ScienceDirect

# LWT

journal homepage: www.elsevier.com/locate/lwt

# Thaw rigor in Atlantic salmon (*Salmo salar*) fillets, as affected by thawing rate and frozen storage time

Bjørn Tore Rotabakk<sup>a,\*</sup>, Lars Helge Stien<sup>b</sup>, Torstein Skåra<sup>a</sup>

<sup>a</sup> Nofima, Norwegian Institute of Food, Fisheries and Aquaculture Research, Richard Johnsens Gate 4, N-4021, Stavanger, Norway
<sup>b</sup> Institute of Marine Research, PO Box 1870 Nordnes, NO-5817, Bergen, Norway

ARTICLE INFO

Keywords: Atlantic salmon Freezing Quality Pre-rigor Fillet contraction

# ABSTRACT

The influence of stress, storage time and thawing regimes was investigated with respect to *rigor* development and other related physical traits, on farmed Atlantic salmon. The fish were sampled from a commercial slaughter-house production line, filleted, vacuum packaged and quickly frozen, *pre-rigor*. The investigated parameters were selected to maintain fresh-like quality.

No significant (p > 0.600) effects were seen between rigor contraction and stress parameters measured at time of slaughter. Frozen storage time significantly (p = 0,003) affected contraction, while no effect of thawing method (p = 0.824) was detected. The liquid loss of the frozen groups was significantly (p = 0.019) affected by thawing method, while frozen storage time had no significant (p = 0.087) effect. Further, drip loss was not affected by fillet contraction (p = 0.075). Increasing frozen storage time significantly (p = 0.042) decreased the redness and thawing in air rather than water significantly (p = 0.015) decreased the yellowness of the fillets. Fillet stored for 1 month had significantly (p = 0.004) lower F-break force, while no significant (p > 0.227) effect of storage or thawing was seen on hardness. Frozen fillets were comparable to 7 days old vacuum packaged fresh fillets stored on ice for seven days, albeit with some increase in drip loss and some differences in texture, but with comparable color and fillet contraction.

#### 1. Introduction

The production of Norwegian farmed salmon has been increasing massively in recent years and in 2019 the volume exceeded 1.1 million tons, with an export value of 6.26 billion GBP (Norwegian Seafood Council, 2020). The bulk part of this volume is exported as fresh on ice, in which the first part of the shelf-life, which is typically 20 days (Sveinsdottir, Ingvarsdottir, Torrissen, Cardinal & Hafsteinsson, 2003), is used for distribution. Airfreight to distant markets has been increasing, but as airfreight is both costly and associated with a high carbon footprint, alternative distribution concepts are sought.

Freezing of fish has a long history, and frozen fish products are widely available, albeit not always associated with high quality and freshness. Recent studies, however, have shown that high raw material quality/freshness, rapid freezing, storage at temperatures below -30 °C and adequate thawing, give rise to quality that can exceed the fresh alternative (Altintzoglou & Heide, 2014, 2016; Altintzoglou, Heide, & Carlehog, 2014; Eikevik, Tolstorebrov, Bantle, Nordtvedt, & Stavset, 2015). One reason for this is that while the quality of fresh fish

deteriorates throughout the shelf life, the degradation can be halted during freezing, and the distribution can take place without compromising quality.

Water is the major component in fish, and it affects several aspects, both regarding yield, but also with respect to sensory characteristics. Farmed Atlantic salmon muscle contains 60–72% water (Aursand, Erikson, & Veliyulin, 2010; Ørnholt-Johansson, Frosch, & Munk Jørgensen, 2017). Muscle foods lose liquid, *post mortem*, due to changed capacity of the muscle to retain its natural water (Huff-Lonergan & Lonergan, 2007).

Liquid loss is also associated with tissue damage due to ice crystal formation during freezing, storage and thawing (Petzold & Aguilera, 2009), which can be perceived as a major obstacle for the quality of frozen food. When freezing, the volume of water increases by approximately 9%, and this expansion, combined with sharp ice-crystals, leads to tissue-damage and degradation (Alizadeh, Chapleau, De Lamballerie, & LeBail, 2007; Benjakul & Bauer, 2001; Wang et al., 2018).

The fat content of Norwegian farmed salmon is approximately 16% (Myhre, Bergejordet, Nordbotten, Løken, & Fagerli, 2012), hence the

\* Corresponding author. Nofima, Norwegian Institute of Food, Fisheries and Aquaculture Research, P.O. Box 8034, N-4021, Stavanger, Norway. *E-mail address:* bjorn.tore.rotabakk@nofima.no (B.T. Rotabakk).

https://doi.org/10.1016/j.lwt.2022.113793

Received 22 April 2022; Received in revised form 17 June 2022; Accepted 18 July 2022 Available online 8 August 2022

0023-6438/© 2022 The Authors. Published by Elsevier Ltd. This is an open access article under the CC BY license (http://creativecommons.org/licenses/by/4.0/).







amount of available water for crystal formation is lower than that of lean fish species. Furthermore, the contraction of fat when freezing, counteracts the damaging effect of expanding crystal formation (Nakazawa & Okazaki, 2020). It is also hypothesized that the distribution of fat in the muscle tissue reduces the free water pools, thus also contributing to limiting the crystal size and subsequent tissue damage.

Raw material freshness is very important for the quality of frozen products, because several degradation processes are initiated *post mortem* that weakens the muscle structure (Delbarre-Ladrat, Cheret, Taylor, & Verrez-Bagnis, 2006). Kaale and Eikevik (2013) found that, during super-chilling, the size of ice crystals formed in pre-rigor muscle was significantly smaller than those formed in post-rigor muscle. Love and Haraldsson (1958) found less cell damage in cod fillets frozen pre rigor, than in those frozen post rigor. Recent studies have also shown that a high pH and high ATP level in fresh fish effectively suppress changes in various properties during frozen storage (Nakazawa & Okazaki, 2020). Optimizing frozen product quality by rapid freezing after slaughter – pre rigor, is feasible in aquaculture. From a production perspective, it is also the best alternative.

For muscle that has been frozen pre-rigor, thaw rigor (McDonald & Jones, 1976) can be seen when the rigor process starts during or after thawing. It can be less strong than in fresh fillets, but still lead to contractions that will affect shape (Misimi, Erikson, Digre, Skavhaug, & Mathiassen, 2008) and potentially also liquid loss. Fillets, which can contract freely without mechanical constraints, are prone to this phenomenon, as shown for cod (Rotabakk, Bleie, Stien, & Roth, 2014).

If quality and yield reduction can be minimized, the benefits of drastically increased shelf life in the frozen state can be utilized for transport and distribution, particularly for distant markets. Stress level, before and at slaughter, is a key factor for the rigor development (Misimi et al., 2008) but there is a lack of data available regarding the effects of stress and handling on thaw rigor and liquid loss for farmed Atlantic salmon. Frozen fish is a very broad category, and the parameters in this study were selected to obtain frozen/thawed products with optimal quality that could be comparable to fresh. Hence the main objective of the present study was to investigate the effect of storage time up to 4 months, and fast and slow thawing rate on the rigor development, and other physical traits (color and texture), on vacuum packaged salmon fillets frozen pre-rigor.

# 2. Materials and methods

#### 2.1. Experimental setup

In January 2020 a total of 25 farmed Atlantic salmon, (4.16  $\pm$  0.70 kg), were sampled during commercial slaughtering in the southwest part of Norway. The fish were collected from the production line after electrical stunning, killed by a sharp blow to the head and individually tagged. Weight, length, muscle pH and blood lactate level were measured prior to bleeding. The fish were then left to bleed in iced water by gill cut, before they were manually gutted, filleted, tagged, and photographed for fillet length, vacuum packed (99%) in OPA/PE bags (130  $\mu$ m, Neemann, Leer, Germany) and frozen in dry ice (-78 °C), prior to storage at – 30 °C. The filleting was carried out *pre rigor*, 1.5–2 h after bleeding.

After frozen storage for one or four months, the left fillet of a fish was thawed rapidly in 4 °C water, while the right fillet was thawed slowly in 4 °C chilled air. In addition, a group stored unfrozen and chilled on ice for 7 days was used as a control. The sampling procedure thus resulted in five groups of salmon fillets: (A) Control (non-frozen – stored on ice/ 0 °C), n = 10, (B) stored frozen, at -30 °C, for one month and thawed rapidly, n = 10, (C) stored frozen for one month and thawed slowly, n = 10, (D) stored frozen for four months and thawed rapidly, n = 10, and (E) stored frozen for four months and thawed slowly, n = 10.

All pouches with frozen fish were opened after thawing for measurement of drip loss and fillet length (photo). After these measurements, the pouches were manually closed by folding, and stored for 7 days at 0  $^{\circ}$ C, prior to new measurements of drip loss and length, and finally also pH, instrumental color, and texture measurements.

The non-frozen fillets were stored, vacuum packaged, for 1 week at 0 °C, prior to measurement of pH, drip loss and length, as well as instrumental color and texture measurements. So due to differences in packaging (air, vacuum) the samples stored for 1 week at 0 °C after thawing cannot be compared directly to the control.

# 2.2. Temperature logging

The temperature was logged in the anterior part of the dorsal muscle, using a 176T4 Testo (Testo, Lenzkrich, Germany) using a Type T insertion probe (Testo Lenzkrich, Germany). Temperature was logged both during freezing and thawing and was measured in duplicate.

#### 2.3. Condition factor

The condition factor (K) was calculated by using Fulton's conditions factor (Formula 1):

$$K = \frac{100 \times W}{L^3} \tag{1}$$

where W = weight (g) and L = the fork length (cm). Condition factor was measured on all whole fish (n = 25).

# 2.4. Blood lactate

Blood samples were extracted from the caudal vein immediately after death from all the sampled individuals. Analysis was performed within 2 min, using a Lactate ProTM 2 blood lactate test meter (Arkray Factory Inc., Shiga, Japan), in combination with the Lactate ProTM 2 test strips (Arkray Factory Inc., Shiga, Japan). Blood lactate were measured on all whole fish (n = 25).

# 2.5. Muscle pH

Muscle pH were measured after bleeding, in the anterior dorsal muscle close to the gills, using a Mettler Toledo SevenGo pro<sup>TM</sup> pH meter (Mettler Toledo Inc, OH, USA) equipped with Inlab puncture electrode. After thawing and storage, muscle pH was measured in the cranial part of the fillet (7 after thawing). pH was measured on all whole fish (n = 25) and all fillets (n = 10) in all groups. Temperature was measured simultaneously to automatically adjust pH according to the sample temperature.

#### 2.6. Dry matter

Dry matter was determined according to (ISO 6496, 1983). Each sample (5.2  $\pm$  0.2 g) was weighed (BP221S, Sartorius, Germany) in preweighed aluminum cups and placed in a drying cabinet (TS8056, Termaks AS, Bergen, Noreat) at 105 °C for 24 h, prior to cooling and reweighing. A total of 11 samples were used for dry matter assessment.

# 2.7. Fillet rigor contraction

Each fillet was photographed (Canon 400D, Canon Inc, Tokyo, Japan) on the day of thawing and after 7 days of chilled storage (n = 10 in each group). The fillets were placed, one-by-one, on a blue background together with a lineal of known length in centimeters and photographed directly from above. The images were later analyzed using a script programmed in Matlab R2020a (MathWorks<sup>TM</sup>) that followed the image analysis procedure described in Stien, Suontama, and Kiessling (2006). In short, for each image the pixels representing the fillet were segmented from the background based on their red color in contrast to

the backgrounds' blue. The length of the fillet region was then calculated in pixels (MajorAxisiLength, function regionprops, Matlab) and converted to centimeters based on the length of the lineal. Fillet contraction after thawing and after 7 day of chilled storage was calculated as percentage difference related to initial fillet length.

# 2.8. Surface color

Fillet surface color (CIE Lab) was assessed on fillets seven days *post mortem* (control) or seven days after thawing by a digital photo imaging color-measuring system (DigiEye full system, VeriVide Ltd., Leicester, UK) (n = 10 in each group). The fillets were placed in a standardized light-box with daylight (6400 K) and photographed with a calibrated digital camera (Nikon D80, 35 mm lens, Nikon Corp., Japan). The pictures were analyzed with DigiPix software (VeriVide Ltd., Leicester, UK) and the color quantified on the loin part of the fillet, (above the centerline and between the Norwegian quality cut and the neck). L\* describes the products lightness (L\* = 100 = white, and L\* = 0 = black), a\* describes the intensity of color on the red–green axis (a\* > 0 = red, and a\* < 0 = green) and b\* the intensity of color on the yellow–blue axis (b\* > 0 = yellow, and b\* < 0 = blue).

# 2.9. Liquid loss

Liquid loss from the fillets (n = 10 in each group) was calculated as the difference in fillet weight between day 0 (before freezing) day 0 (after thawing) and day 7 (after storage of fresh (control) or thawed fillet at 0 °C).

#### $LL = [(m_0 - m_x)/m_0] * 100\%$

where  $m_0 = \text{initial}$  fillet weight at day 0, before freezing.  $m_x = \text{fillet}$  weight at day x.

#### 2.10. Texture

Instrumental textural analyses were performed using a Texture Analyser TA-XT2 (SMS Ltd., Surrey, England) equipped with a 25 kg load cell (modified method after Lerfall et al. (2012)). A flat-ended cylinder probe (20 mm diameter, type P/1SP) was used. The force-time graph was recorded by a computer equipped with the Texture Exponent light software for windows (version 4.13, SMS). This software was also used to analyze the data. Analyses were performed in triplicates (average values were used in data analysis) of each fillet (n = 10 in each group) 7 days *post mortem* (control) or after thawing. The resistance force (N) was recorded with a constant speed of 5 mm s<sup>-1</sup>, and the force required to penetrate the surface was recorded as breaking force (F-break), and the force required to press the cylinder down to 60% of fillet thickness was used to describe the textural parameter fillet hardness.

#### 2.11. Statistics

Statistical analysis included regression, analysis of variance (one way-ANOVA), general linear model (GLM) and Tukey's HSD test (p < 0.05), and was done in Minitab v 19 (Minitab, USA). Values are represented as mean  $\pm$  SD, unless other stated.

# 3. Results and discussion

The main objective of the present study was to investigate the influence of storage times and thawing regimes on the rigor development, and other physical traits (color and texture), on salmon fillets frozen prerigor. The control was not frozen but vacuum packaged pre rigor. Veiseth-Kent et al. (2010) found pre rigor vacuum packaged salmon fillets to have less favorable quality traits than those of fillets packaged after contraction or filleted post rigor. From a production and distribution perspective, however, immediate packaging after filleting makes sense, and hence this combination was chosen. Veiseth-Kent et al. (2010) saw indications of that the effects seen on the pre-rigor packaged salmon fillets could be related to that their contraction was restricted during rigor development. In this experiment, however, some release of rigor could be expected to take place during thawing, and hence the control should be vacuum packaged. In addition, comparing newly thawed with one-week fresh fillets is relevant from a commercial point of view, as this represents two natural options regarding quality.

#### 3.1. Composition

Based on the dry matter content determinations, the water content was  $62.3 \pm 0.9\%$  (n = 11), well within the range reported by Aursand et al. (2010). Fullers condition factor for the group was  $1.42 \pm 0.11$ , which is in the high end of the range considered as normal (0.9–1.5) (Stien et al., 2013).

# 3.2. Freezing and thawing

During freezing, the time to reach a core temperature below -30 °C was measured to be less than 30 min (Fig. 1). During thawing, reaching a core temperature of 1.5 °C was measured to take approx. 4 h when thawing in water, while thawing in air took approx. 12 h.

#### 3.3. Stress parameters

In this study commercially harvested and stunned fish were sampled directly from a commercial production line, and the pH and the blood lactate level in the salmon muscle were measured, in order to have some indication of the stress level of the fish. The muscle pH was in average  $6.58 \pm 0.18$  and the blood lactate level was  $6.3 \pm 2.9$  mmol/L. Lerfall et al. (2015) measured the muscle pH of uncrowded fish (from sea cage, control) and fish that were crowded, pumped and live chilled (stressed) to  $7.35 \pm 0.09$  and  $6.72 \pm 0.14$ , respectively, and the corresponding lactate values to  $0.37 \pm 0.16$  and  $7.23 \pm 1.15$  mmol/L. In their experiment, Einen, Guerin, Fjaera, and Skjervold (2002) measured pH in *pre-rigor* filleted salmon to 6.6 at the time of filleting. Our results suggest that most fish were stressed as can be expected from commercial handling, and a weak, but significant (p < 0.001) correlation (R<sup>2</sup> = 43.4%) between the two parameters was seen in the measured intervals (Fig. 2).

pH was measured again after 7 days of chilled storage. Frozen storage time (p < 0.001), thawing method (p = 0.010) and the interaction between them (p = 0.008) (Table 1) showed significant effects on pH. Considering the 4 groups, however, only fillets stored frozen for 4 months and thawed fast in water showed a significantly (p < 0.001) decreased pH compared to the other three groups. The control group had a pH of  $6.26 \pm 0.064$  after 7 days of chilled storage in vacuum. pH after 7 days is comparable to other results, where Lerfall et al. (2015) found a pH of approx. 6.25 in fresh *pre* rigor fillets after 4 days of storage, while frozen and thawed salmon fillets had a pH of approx. 6.2 (Einen et al., 2002).

# 3.4. Contraction

All fish go through a *rigor* state after slaughtering. During *rigor*, the acto-myosin complex become irreversible connected, and the fillet shrinks. Thaw rigor is a muscle contraction that occurs by utilizing the energy of residual ATP during thawing (Nakazawa & Okazaki, 2020). No significant (p = 0.824) effect of thawing method was detected on the length contraction, but there was a significant effect (p = 0,003) of frozen storage time. Fillets thawed after one month of frozen storage, had an average contraction of  $8.57 \pm 2.48\%$ , while the fillets stored for 4 months had an average contraction of  $5.97 \pm 2.57\%$ . In addition,

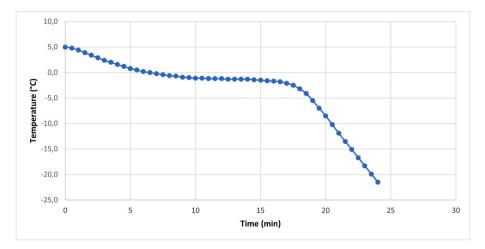


Fig. 1. Temperature measured in the thermal center of a salmon fillet during freezing in contact with dry ice (solid CO<sub>2</sub>.). The figure is from one representative measurement.

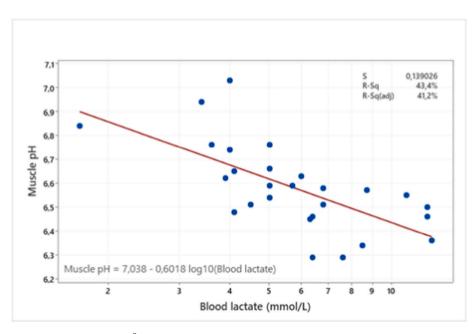


Fig. 2. Log-linear regression ((p < 0.001) correlation ( $R^2 = 43.4\%$ )) between Muscle pH and blood Lactate (mmol/L) in Atlantic salmon, after crowding, pumping and stunning. Blood lactate was log transformed.

Table 1

pH measured in pre rigor filleted Atlantic salmon fillets, frozen in vacuum packages, thawed and stored chilled (0 °C) in open plastic bags for 7 days. Superscript letters indicate statistically significant(p < 0.001) differences. Number denotes number of months stored frozen, while "A" denotes thawed slowly in air (12 h, 4 °C), and "W" denotes thawed fast in water (4 h, 4 °C).

Group	pH
1 W	$6.32\pm0.059~^a$
1 A	$6.31\pm0.12~^{\rm a}$
4 W	$6.12 \pm 0.072 \ ^{\rm b}$
4 A	$6.27\pm0.077~^{a}$

one-week storage after thawing significantly (p < 0.001) affected the magnitude of contraction, where the fillets contracted an additional 1.78  $\pm$  2.25%. No interaction effect (p = 0.965) between frozen storage

time and thawed storage time was detected. In comparison, the fresh control had an average contraction of 7.63  $\pm$  2.13% after 7 days of chilled storage in vacuum. Only fillets stored frozen for one month and stored chilled for 7 days were significantly (p < 0.05) different from the control, with an average contraction of 10.59  $\pm$  2.36%. This is in contradiction to the findings of Einen et al. (2002). They found almost ignorable contraction (0,5-2% in length) in pre-rigor fillets that had been quickly frozen, and stored for 4 days at -25 °C prior to slow thawing in air (4 °C, 30 h), compared to the approx. 10% contraction in the pre-rigor control fillets at 4 °C. They suggested that the remaining ATP in the muscle would be degraded at low temperatures (sub-zero) during the slow thawing, and insufficient amounts be left for muscular contraction. It is known that degradation of ATP in frozen fish occurs as low as -40 °C (Li et al., 2019), but our study did not show any significant effect of thawing rate. However, degradation of ATP during frozen storage can explain the decreased fillet contraction observed for the fillets stored for 4 months compared to 1 month. Further, it seems like not all ATP and glycogen was consumed during thawing, as the fillets continued to contract during the 7 days of chilled storage. This could also be related

to the fact that the chilled storage took place in opened packages, without restriction of contraction (Veiseth-Kent et al., 2010).

The stress parameters were recorded with the intention to be correlated to rigor contraction, as shown previously on salmon (Veiseth, Fjæra, Bjerkeng, & Skjervold, 2006) and Atlantic cod (*Gadus morhua*) (Jørpeland, Imsland, Stien, Bleie, & Roth, 2013). However, no significant (p > 0.600) effect between rigor contraction and stress parameters were found when including lactate and pH at time of slaughter as covariates in the ANOVA test.

# 3.5. Liquid loss

Including lactate level at slaughtering as a covariate into the GLM did not contribute significantly (p = 0.584) to the explanation of the observed variance, when studying the liquid loss after thawing. However, when including initial pH, a significant (r = 0.220, p = 0,001) increase in liquid loss could be observed with increasing initial pH. The level of *pre-mortem* stress has not been found to have a significant effect on drip loss in salmon fillets (Lerfall et al., 2015; Roth, Birkeland, & Oyarzun, 2009), so the observed correlation between initial pH and drip loss in the present study might be spurious.

The liquid loss was significantly (p = 0.019) affected by thawing method, where thawing in air gave an average liquid loss of 0.94  $\pm$ 0.24% compared to 0.81  $\pm$  0.19% after thawing in water. Similar results were seen when thawing rainbow trout (Oncorhynchus mykiss) (Javadian, Rezaei, Soltani, Kazemian, & Pourgholam, 2013), chicken breast (Oliveira, Gubert, Roman, Kempka, & Prestes, 2015). Decreased drip loss after rapid thawing in water might be due to a fast process that leads to a shorter phase transition time, and by that less cell damage (Stormo & Skara, 2021). The control group had an average liquid loss of 0.80  $\pm$ 0.08% after 7 days. This is comparable to the thawing loss in all the frozen samples, and no significant (p < 0.05) difference was detected between the control and the thawed samples. This shows that transport and storage of frozen salmon has a potential to provide a product with comparable quality as 7 days old non-frozen salmon, when comparing drip loss. Frozen storage time did not have a significant (p = 0.087) impact on the liquid loss. In addition, no significant interaction (p = 0.097) was found between thawing method and storage time. However, samples stored for 4 months and thawed in air had a significantly (p < p

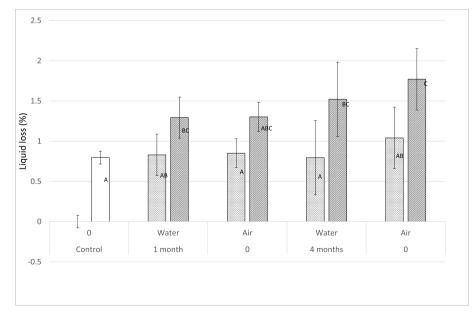
0.05) increased liquid loss compared to both storage times thawed in water (Fig. 3) when comparing the four groups at the day of thawing.

Muscle degradation as an effect of storage time has been related to increased drip loss (Ofstad et al., 1996), and a significantly (p < 0.001) increased drip loss was observed for all groups after 7 days of chilled storage (Fig. 3). Frozen storage time (p = 0.001) had a significant impact on the increase of liquid loss, where 1 month storage and 4 months gave increased liquid loss of 0.46  $\pm$  0.22% and 0.74  $\pm$  0.26%, respectively. Temperature fluctuations is known to affect size and distribution of ice crystals that damage the food quality (Zhao & Takhar, 2017) and are inevitable even when samples are stored at  $-30~^\circ\text{C}$  in commercial freezers. This might explain the observed increased drip loss in samples stored frozen for 4 months. Thawing method on the other hand had no significant effect (p = 0.939) on the drip loss. In addition, no significant (p = 0.831) interaction between frozen storage time and thawing method was detected.

Muscle shortening during *rigor* has been related to increased liquid loss (Huff-Lonergan & Lonergan, 2005), and in the present study, a significant (p = 0.035) but very weak ( $R^2 = 5.4\%$ ) positive correlation was found, where increased contraction gave increased liquid loss (LQ % = 0.835 + 0.0350 Contraction %) in the frozen samples. However, by including frozen and chilled storage time as categorical predictors, the correlation between contraction and liquid loss became insignificant (p = 0.075).

#### 3.6. Color

Color is one of the most important quality traits of salmon, and instrumental color analyses is a rapid and objective method for determination of color in salmonid muscle (fillets). It correlates to both carotenoid concentration and sensory color assessments (Skrede, Risvik, Huber, Enersen, & Blumlein, 1990; Skrede & Storebakken, 1986a, 1986b). Several studies have documented that the color is affected by storage (Ottestad, Sørheim, Heia, Skaret, & Wold, 2011), packaging (Einen et al., 2002; Ottestad et al., 2011) and freezing (Einen et al., 2002). The color of the vacuum packaged control samples, L\*: 52.58, a\*: 37.36 and b\*: 33.49 was measured after one week storage at 0 °C. These values correspond reasonably well to those found by Ottestad et al. (2011) in vacuum packaged salmon stored for 6 days at 2 °C: 48.7, 41.2



**Fig. 3.** Liquid loss of salmon fillets (means  $\pm$  st.dev), stored frozen for 1 or 4 months at – 30 °C, and thawed rapidly in water (4 °C, 4 h) or slowly in air (4 °C, 12 h). Light grey bars represent liquid loss directly after thawing, while dark grey bars represent liquid loss after 7 days of chilled storage (0.1 °C). White bar represents liquid loss for the fresh and vacuum packaged control, after 7 days of chilled storage (0.1 °C). n  $\geq$  8.

and 44.6, respectively. As in our experiment, these values were determined by imaging, which is important to bear in mind as they were shown to have significantly higher values than those obtained with designated color measurement instrumentation (e.g. Minolta).

When studying the frozen and thawed samples, increasing frozen storage from 1 to 4 months significantly (p = 0.042) decreased the a<sup>\*</sup>values from 35.94  $\pm$  1.88 to 34.58  $\pm$  2.16. In addition, thawing in air compared to water significantly (p = 0.015) decreased the b\*-values from 33.30  $\pm$  1.55 to 31.99  $\pm$  1.68. No other significant (p > 0.330) main or interaction effects on a\* or b\* were detected. When comparing the frozen and thawed fillets with the control, the fillets stored for 4 months had a significantly (p = 0.007) decreased a\*-value, and the fillets thawed in air had a significantly (p = 0.034) decreased b\*-value. No other significant (p < 0.005) differences were found. In addition, none of the factors (frozen storage time or thawing method) affected the L\* significantly (p > 0.220), including the interaction between them. This differs from the findings of Rørå and Einen (2003), that freezing increased the lightness and the color intensity of the fillets. But the frozen storage period in that study was only 6 days, and the description of the packaging is limited. Carotenoids may be oxidized by freezing and thawing of Atlantic salmon (Regost, Jakobsen, & Rørå, 2004; Sheehan, O'Connor, Sheehy, Buckley, & FitzGerald, 1998), which can explain the observed decrease in a\*-value by increased frozen storage time. However, the observed color effects were small in practical terms.

# 3.7. Texture

In addition to color, texture is among the most important parameters that determine the overall quality perception of salmon products (Koteng, 1992). Mørkøre and Einen (2003) found several response variables derived from instrumental texture analysis, that correlated with sensory evaluated hardness.

The texture profile analysis (Table 2) revealed differences in the breaking force (F-break) where storage for 1 month had significantly (p = 0.004) lower F-break (18.09  $\pm$  3.20 N) compared to storage for 4 months (25.59  $\pm$  13.51 N). The height was measured simultaneously as the F-break, and the fillets stored for 1 month were significantly higher (p < 0.001) than those stored for 4 months, with a height of 26.55  $\pm$ 3.19 mm and 23.70  $\pm$  3.29 mm, respectively. Fillet height was for that reason tested as a covariate, but no significant (p = 0.211) effect of fillet height on F-break was detected, and hence it was excluded from the ANOVA. In addition, no other significant (p < 0.141) main or interaction effects were detected. These results correspond very well, also, with the fillet contraction which was highest after one-month frozen storage. Similar correlations between fillet height and contraction were seen by Veiseth-Kent et al. (2010), who found a significant increased fillet height on fillets filleted post-rigor compared to pre-rigor fillets that were restricted from contraction.

When studying the hardness, no significant (p > 0.227) main or interaction effects were detected between the frozen samples.

Comparing the four groups with the control, shows that samples stored for four months and thawed in air was the only group having a significantly (p < 0.000) tougher surface than the control (Table 2). In addition, the same group was the only group that were not found significantly (p > 0.05) softer that the control when studying the hardness. Sigurgisladottir, Ingvarsdottir, Torrissen, Cardinal, and Hafsteinsson (2000) observed a significant decreased shear force in frozen fish, while Regost et al. (2004) found a softer fillet after frozen storage of fresh salmon fillets when compressed to 60% of the fillet height. This is comparable to the result found in this study. Einen et al. (2002), on the other hand, saw a significant decrease in break force in frozen samples, where the present study found increased or no difference. It has been shown that the structure of whole fresh muscle changes during freezing and frozen storage due to movement of water to extracellular spaces (Sigurgisladottir et al., 2000), which further is affected by freezing rate (Tan, Mei, & Xie, 2021) and temperature fluctuations during storage

#### Table 2

Compression force (N) required to penetrate a 20 mm cylindrical flat ended probe to 60% of the initial height. Tukeys comparison p = 0.05. Number denotes number of months stored frozen at – 30 °C, while "A" denotes thawed slowly in air (12 h, 4 °C), and "W" denotes thawed fast in water (4 h, 4 °C).

Group	F-break	Juiciness
1 W	$17.91\pm3.50^{\rm a}$	$22.49 \pm \mathbf{4.38^a}$
1 A	$18.27\pm2.92^{\rm a}$	$22.39\pm2.77^{\rm a}$
4 W	$23.13\pm11.01^{\rm ab}$	$23.11\pm3.43^{\rm a}$
4 A	$28.04\pm15.40^{\rm b}$	$24.16\pm5.76^{ab}$
Control	$19.63\pm2.85^{\rm a}$	$26.23\pm4.38^{\rm b}$
p-value	<0.000	0.004

#### (Zhao & Takhar, 2017).

#### 4. Concluding remarks

In summary our results indicate that vacuum packaged, pre rigor frozen salmon fillets possess quality traits that are comparable to vacuum packaged salmon fillets stored chilled for seven days. The liquid loss is low (<1%), and somewhat correlated to contraction, but the correlation is weak. After one month no differences in color can be detected between frozen thawed and fresh fillets. Frozen storage for four months gave rise to a slight decrease in redness, but with rapid thawing this remains the only significant difference to fresh. In addition, freezing gave an overall decreased hardness compared to the fresh fillet, while breaking force were generally comparable. One to two months of frozen storage is sufficient to reach distant salmon markets, and with adequate traceability systems and procedures for distribution, our results indicate that salmon fillets of similar quality to fresh, can be presented.

#### Founding

This work was supported by The Research Council of Norway (grant number 194050). The founding source was not involved in conducting the research and preparation of the paper.

# CRediT authorship contribution statement

**Bjørn Tore Rotabakk:** Conceptualization, Formal analysis, Data curation, Writing – original draft. **Lars Helge Stien:** Methodology, Formal analysis, Writing – original draft. **Torstein Skåra:** Conceptualization, Formal analysis, Data curation, Investigation, Writing – original draft, Project administration, Funding acquisition.

#### Declaration of competing interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests: Torstein Skåra and Bjørn Tore Rotabakk reports financial support was provided by The research council of Norway.

#### References

- Einen et al. (2002) is one of the first papers describing freezing of *pre-rigor* Atlantic salmon. They compared *pre* and *post-rigor* freezing, related to drip-loss, texture, gaping color and rigor contraction, which gives an important basis for our study.
- Kaale and Eikevik (2013) study the formation of ice crystals in Atlantic salmon during superchilling and stuperchilled storage. They documented the effect of slow and fast superchilling, and concluded that fast chilling/partly freezing gives small crystals.
- Sigurgisladottir et al. (2000) studied the effects of freezing and thawing on the muscle fibers of Atlantic salmon, and provide knowledge to understand the results of our study.
- Stien et al. (2006) provides the method for using images for studying fillet contraction. This method was vital in the present study.
- Lerfall et al. (2015) studyed the effect of *pre mortem* stress on the quality of *pre-rigor* filleted Atlantic salmon. Eventhough they did not study frozen salmon, this paper enlighted the effect of stress on quality.

#### B.T. Rotabakk et al.

Alizadeh, E., Chapleau, N., De Lamballerie, M., & LeBail, A. (2007). Effects of freezing and thawing processes on the quality of atlantic salmon (salmo salar) fillets. *Journal* of Food Science, 72(5), E279–E284.

Altintzoglou, T., & Heide, M. (2014). A comparison of French and English consumers' expectations regarding labeled cod fillet products. *Journal of Aquatic Food Product Technology*, 23(2), 155–164.

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(6), 885–894.

Altintzoglou, T., Heide, M., & Carlehog, M. (2014). French consumer profiles' reactions to information on cod fillet products. *British Food Journal*, 116(3), 374–389.

Aursand, I. G., Erikson, U., & Veliyulin, E. (2010). Water properties and salt uptake in atlantic salmon fillets as affected by ante-mortem stress, rigor mortis, and brine salting: A low-field 1H NMR and 1H/23Na MRI study. *Food Chemistry*, 120(2), 482–489.

Benjakul, S., & Bauer, F. (2001). Biochemical and physicochemical changes in catfish (silurus glanis linne) muscle as influenced by different freeze-thaw cycles. *Food Chemistry*, 72(2), 207–217.

Delbarre-Ladrat, C., Cheret, R., Taylor, R., & Verrez-Bagnis, V. (2006). Trends in postmortem aging in fish: Understanding of proteolysis and disorganization of the myofibrillar structure. *Critical Reviews in Food Science and Nutrition*, 46(5), 409–421.

Eikevik, T. M., Tolstorebrov, I., Bantle, M., Nordtvedt, T. S., & Stavset, O. (2015). Challenges of the usage of ultra-low temperatures for fish freezing and storage. In Proceedings of the 24th international congress of refrigeration, ISBN 978-2-36215-012-8.

\* Einen, O., Guerin, T., Fjaera, S. O., & Skjervold, P. O. (2002). Freezing of pre-rigor fillets of atlantic salmon. *Aquaculture*, 212(1–4), 129–140.

Huff-Lonergan, E., & Lonergan, S. M. (2005). Mechanisms of water-holding capacity of meat: The role of postmortem biochemical and structural changes. *Meat Science*, 71 (1), 194–204.

Huff-Lonergan, E., & Lonergan, S. M. (2007). New frontiers in understanding drip loss in pork: Recent insights on the role of postmortem muscle biochemistry. *Journal of Animal Breeding and Genetics*, 124, 19–26.

Javadian, S. R., Rezaei, M., Soltani, M., Kazemian, M., & Pourgholam, R. (2013). Effects of thawing methods on chemical, biochemical, and microbial quality of frozen whole rainbow trout (oncorhynchus mykiss). *Journal of Aquatic Food Product Technology*, 22 (2), 168–177.

Jørpeland, G., Imsland, A., Stien, L. H., Bleie, H., & Roth, B. (2013). Effects of filleting method, stress, storage and season on the quality of farmed atlantic cod (gadus morhua l.). Aquaculture Research, 1–11.

\*\* Kaale, L. D., & Eikevik, T. M. (2013). A study of the ice crystal sizes of red muscle of pre-rigor atlantic salmon (salmo salar) fillets during superchilled storage. *Journal of Food Engeneering*, 119(3), 544–551.

Koteng, A. (1992). Markedsundersøkelse norsk laks (market investigation Norwegian salmon). Fiskerinæringens Landsforening (FHL).

Lerfall, J., Larsson, T., Birkeland, S., Taksdal, T., Dalgaard, P., Afanasyev, S., et al. (2012). Effect of pancreas disease (pd) on quality attributes of raw and smoked fillets of atlantic salmon (salmo salar l.). Aquaculture, 324, 209–217.

\*\*\* Lerfall, J., Roth, B., Skare, E. F., Henriksen, A., Betten, T., Dziatkowiak-Stefaniak, M. A., et al. (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.

Li, D. P., Qin, N., Zhang, L. T., Li, Q., Prinyawiwatkul, W., & Luo, Y. K. (2019). Degradation of adenosine triphosphate, water loss and textural changes in frozen common carp (cyprinus carpio) fillets during storage at different temperatures. International Journal of Refrigeration-Revue Internationale Du Froid, 98, 294–301.

Love, R. M., & Haraldsson, S. B. (1958). Thaw rigor and cell rupture. *Nature*, 181(4619), 1334-1334.
McDonald, I., & Jones, N. R. (1976). Control of thaw rigor by manipulation of

temperature in cold store. Journal of Food Technology, 11(1), 69–71.

Misimi, E., Erikson, U., Digre, H., Skavhaug, A., & Mathiassen, J. R. (2008). Computer vision-based evaluation of pre- and postrigor changes in size and shape of atlantic cod (gadus morhua) and atlantic salmon (salmo salar) fillets during rigor mortis and ice storage: Effects of perimortem handling stress. *Journal of Food Science*, 73(2), E57–E68.

Mørkøre, T., & Einen, O. (2003). Relating sensory and instrumental texture analyses of atlantic salmon. *Journal of Food Science*, 68(4), 1492–1497.

Myhre, J. B., Bergejordet, Å., Nordbotten, A., Løken, E. B., & Fagerli, R. A. (2012). Nutritional composition of selected wild and farmed raw fish. Oslo: Mattilsynet. Nakazawa, N., & Okazaki, E. (2020). Recent research on factors influencing the quality of

frozen seafood. Fisheries Science, 86, 231–244.

Norwegian Seafood Council. (2020). Sjømateksport for 107,3 milliarder kroner i 2019. Received from https://seafood.no/aktuelt/nyheter/sjomateksport-for-1073-milliar der-kroner-i-2019/ Accessed 2020 10.6.2020.

Ofstad, R., Egelandsdal, B., Kidman, S., Myklebust, R., Olsen, R. L., & Hermansson, A. M. (1996). Liquid loss as effected by post mortem ultrastructural changes in fish muscle: Cod (gadus morhua l) and salmon (salmo salar). *Journal of the Science of Food and Agriculture*, 71(3), 301–312.

Oliveira, M. R., Gubert, G., Roman, S. S., Kempka, A. P., & Prestes, R. C. (2015). Meat quality of chicken breast subjected to different thawing methods. *Brazilian Journal of Poultry Science*, 17(2), 165–171.

Ørnholt-Johansson, G., Frosch, S., & Munk Jørgensen, B. (2017). Variation in some quality attributes of Atlantic salmon fillets from aquaculture related to geographic origin and water temperature. Aquaculture, 479, 378–383.

Ottestad, S., Sørheim, O., Heia, K., Skaret, J., & Wold, J. P. (2011). Effects of storage atmosphere and heme state on the color and visible reflectance spectra of salmon (salmo salar) fillets. *Journal of Agricultural and Food Chemistry*, 59(14), 7825–7831.

Petzold, G., & Aguilera, J. (2009). Ice morphology: Fundamentals and technological applications in foods. *Food Biophysics*, 4, 378–396.

Regost, C., Jakobsen, J. V., & Rørå, A. M. B. (2004). Flesh quality of raw and smoked fillets of atlantic salmon as influenced by dietary oil sources and frozen storage. *Food Research International*, 37(3), 259–271.

Rørå, A. M. B., & Einen, O. (2003). Effects of freezing on quality of cold-smoked salmon based on measurements of physiochemical characteristics. *Journal of Food Science*, 68 (6), 2123–2128.

Rotabakk, B. T., Bleie, H., Stien, L. H., & Roth, B. (2014). Effect of blood removal protocol and superchilling on quality parameters of prerigor filleted farmed atlantic cod (gadus morhua). *Journal of Food Science*, 79(5), E881–E886.

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(3–4), 350–356.

Sheehan, E. M., O'Connor, T. P., Sheehy, P. J. A., Buckley, D. J., & FitzGerald, R. (1998). Stability of astaxanthin and canthaxanthin in raw and smoked atlantic salmon (salmo salar) during frozen storage. *Food Chemistry*, 63(3), 313–317.

\*\*\*\* Sigurgisladottir, S., Ingvarsdottir, H., Torrissen, O. J., Cardinal, M., & Hafsteinsson, H. (2000). Effects of freezing/thawing on the microstructure and the texture of smoked atlantic salmon (salmo salar). Food Research International. 33(10). 857–865.

Skrede, G., Risvik, E., Huber, M., Enersen, G., & Blumlein, L. (1990). Developing a color card for raw flesh of astaxanthin-fed salmon. *Journal of Food Science*, 55(2), 361–363.

Skrede, G., & Storebakken, T. (1986a). Characteristics of color in raw, baked and smoked wild and pen-reared atlantic salmon. *Journal of Food Science*, 51(3), 804–808.

Skrede, G., & Storebakken, T. (1986b). Instrumental color analysis of farmed and wild atlantic salmon when raw, baked and smoked. *Aquaculture*, 53(3–4), 279–286.

Stien, L. H., Bracke, M. B. M., Folkedal, O., Nilsson, J., Oppedal, F., Torgersen, T., et al. (2013). Salmon Welfare index model (SWIM 1.0): A semantic model for overall welfare assessment of caged atlantic salmon: Review of the selected welfare indicators and model presentation. *Reviews in Aquaculture*, 5(1), 33–57.

\*\*\*\*\* Stien, L. H., Suontama, J., & Kiessling, A. (2006). Image analysis as a tool to quantify rigor contraction in pre-rigor-filleted fillets. *Computers and Electronics in Agriculture*, 50(2), 109–120.

Stormo, S. K., & Skara, T. (2021). Liquid loss in thawed cod-deconvoluting the effects of freezing-rate, freezing cycles, frozen storage time, and thawing-rate through a full factorial design. *Journal of Food Process Engineering*, 44(6).

Sveinsdottir, K., Hyldig, G., Martinsdottir, E., Jörgensen, B., & Kristbergsson, K. (2003). Quality index method (qim) scheme developed for farmed atlantic salmon (salmo salar). Food Quality and Preference, 14(3), 237–245.

Tan, M. T., Mei, J., & Xie, J. (2021). the formation and control of ice crystal and its impact on the quality of frozen aquatic products: A review. *Crystals*, 11(1).

Veiseth-Kent, E., Hildrum, J. I., Ofstad, R., Rora, M. B., Lea, P., & Rodbotten, M. (2010). The effect of postmortem processing treatments on quality attributes of raw atlantic salmon (salmo salar) measured by sensory and instrumental methods. *Food Chemistry*, 121(1), 275–281.

Veiseth, E., Fjæra, S. O., Bjerkeng, B., & Skjervold, P. O. (2006). Accelerated recovery of atlantic salmon (salmo salar) from effects of crowding by swimming. *Comparative Biochemistry and Physiology Part B: Biochemistry and Molecular Biology*, 144(3), 351–358.

Wang, Y., Miyazaki, R., Saitou, S., Hirasaka, K., Takeshita, S., Tachibana, K., et al. (2018). The effect of ice crystals formations on the flesh quality of frozen horse mackerel (trachurus japonicus). *Journal of Texture Studies*, 49(5), 485–491.

Zhao, Y., & Takhar, P. S. (2017). Freezing of foods: Mathematical and experimental aspects. Food Engineering Reviews, 9(1), 1–12.