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Full Length Research Paper

The biochemical textural and sensory properties of frozen stored (-22°C) king scallop (*Pecten maxinus*) meats

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Post-rigor king scallop meats (Pecten maximus) were frozen individually for 24 h at -80°C and kept vacuum packed at -22°C for up to 301 days. Sampling was carried out on fresh meats and at days 1, 28, 91, 154, 210 and 301 of frozen storage. Tests related to muscle integrity (β-hydroxy-acyl-coenzyme –A dehydrogenase activities in muscle extracts), myofibrillar protein denaturation (Ca^{2+}-ATPase activities in actomyosin extracts) showed that storage time affected the integrity of muscles and caused structural changes to myosin (or 'actomyosin'). The water holding capacity (expressible fluids) of the frozen stored scallop meats decreased with storage time and was associated with the denaturation of myofibrillar proteins. The peak shear forces of frozen stored scallop meats, as measured by the Warner-Bratzler shear knife, did not change with storage time, indicating that the length of time of storage at -22°C did not affect the tenderness of the raw scallop meats. The storage time affected the sensory attributes (flavour, texture and acceptability) of the frozen scallop meats, but these products were in acceptable eating condition after a storage period of ten months. Ca^{2+}-ATPase activities in actomyosin extracts may be useful for assessing the quality loss of the frozen stored scallop meats, since a good linear correlation was obtained with storage time and scores of sensory attributes.

Key words: Scallop meats, frozen storage, quality, enzymes.

INTRODUCTION

Fresh fish are highly perishable commodities and are stored in ice until sold. The need for freezing of fish arises when preservation by chilling with ice is unsuitable for the period of storage time involved (FAO, 1977). However, freezing and storage of frozen fish may furnish favourable conditions for alterations in muscle structure, muscle proteins and lipids and textural properties in general. These changes are related to alterations in the sensory attributes of frozen fish and may affect their market (Shenouda, 1980; Mackie, 1993).

Formation and growth of ice crystals during freezing and storage of fish may cause lysis of organelles and disintegration of membranes. Enzymes that are released from organelles and/or membranes may be more active than in the bound state (Hultin, 1985). The release of dehydrogenases from mitochondria might influence the redox potential of tissues and the release of lipases from lysosomes may cause more rapid breakdown of lipids (Shewfelt, 1981; Civera et al., 1996). The damage of organelles due to freezing and during storage can be studied by the activities of enzymes in muscle tissue fluids, enzymes that in fresh tissue are retained in intracellular organelles. The activity of the mitochondrial enzyme β-hydroxy-acyl-coenzyme A- dehydrogenase (*β*-HAD H) has been regarded as a measure of the damage caused in mitochondria by various freezing and thawing treatments of meat and fish products (Gottesman and Hamm, 1983; Makri et al., 2007).

Lipids that are liberated from the disintegrated membranes could be subjected to faster hydrolytic or oxidative reactions. The secondary oxidation products of lipids influence directly the flavour or interact with proteins and

Abbreviations: **HADH**, β-Hydroxy-acyl-coenzyme A dehydrogenase activity; **TBARS**, thiobarbituric acid reactive substances; **TCA**, trichloroacetic acid.

cause off colours. The changes in lipids during storage of frozen fish can be detected by a variety of chemical tests including free fatty acids content, peroxide value, conjugated dines, thiobarbituric reactive substances (TBARS) etc (Shenouda, 1980).

Changes in the moisture phase during freezing and frozen storage of fish may provide an environment that is conductive to changes in the myofibrillar proteins (Mackie, 1993). Alterations in myofibrillar proteins have been associated with the changes in water holding capacity and textural properties of frozen stored fish. The changes in myofibrillar proteins can be detected in the form of reduced solubility and extractability in saline and other extracting solutions and also reductions in myosin and actomyosin ATP-ase activities, sulfhydryl groups, apparent viscosity, gel-forming ability etc (Shenouda, 1980).

The physical, chemical, bio-chemical and sensory properties of frozen stored fish species have been studied for several decades because of their economic importance. However, little information is available on the quality of frozen stored bivalve molluscs that comprise a significant marine resource.

King scallop (*Pecten maximus*), a bivalve mollusc, is widely distributed in northwest Europe (Brand, 1991). It is much prized as food and the adductor meat, the major edible part of king scallop, is offered to the consumers mainly as fresh and frozen product (Hardy and Smith, 1986). In addition, the exports of king scallops from the producer countries are based mostly on frozen products. For instance, the production of frozen scallops in U.K. in the year 2004 accounted for 20% of the total production of scallops (FAO: http://faostat.fao.org). Freezing, therefore, forms an important section of scallops industry.

There are some reports in the literature on freezing and cold storage of scallop species (Dyer and Hiltz, 1974; Aurell et al., 1976; Chung and Merritt, 1991a, b; Kawashima and Yamanaka, 1992, 1995 a,b; 1996), but there is a lack of information on the quality of frozen King scallop adductor meat. Therefore, the stability in storage of king scallop adductor meat need to be investigated.

The present study is aimed to investigate the effects of the length of time in frozen storage on the quality of scallop meats (*P. maximus*) in regard to the integrity of muscle structure, myofibrillar protein denaturation, lipid degradation, instrumental texture and sensory changes. The information obtained would be useful for assessing the quality of king scallop meats during storage for commer-cial purposes.

MATERIALS AND METHODS

Scallop meats processing and storage

A total number of 105 whole scallops (*P. maximus*), from the Orkney fishing area, were purchased from the Aberdeen fish market. The whole live scallops were delivered to the School of Life Sciences of the Robert Gordon University packed in crushed ice on the same day as their arrival at the fish market.

At the laboratory, the scallops were shucked and the striated part of the adductor muscle, that is, the scallop meat, was separated from all other tissues. In order to ensure that the scallop meats were in post rigor state, they were stored in glass jars without any washing, buried in crushed ice and stored in a chill room at 2 to 4°C for 3 days. Post-rigor scallop meats were required in order to ensure that any difference in instrumental texture measurements would indeed result from differences in freezing and frozen storage and were not due to the development of rigor in the raw scallop muscles prior to freezing. The mean weight of scallop meats was 35 ± 5.6 g (mean ± S.D.).

At the end of the storage period, the scallop meats were divided into 7 groups, each containing 15 scallop meats. The scallop meats from the first group were analyzed as unfrozen controls and the others were individually frozen at -80°C for 24 h on perforated stainless steel sheets. During freezing, the temperature of the centre of 6 scallop meats was monitored using T -type thermocouples and a recording thermometer (Comark Instruments, U.K.). Immediately after freezing, the scallop meats were placed in polyethylene food bags, each containing 5 frozen scallop meats. The bags were vacuum-packed and stored in a domestic freezer cabinet at -22°C.

The stored frozen scallop meats were analyzed for physical, biochemical and sensory properties immediately after freezing and after 28, 91, 154, 210 and 301 days in frozen storage. In each sampling period, 15 frozen scallop meats, that is, the content of 3 bags, were analyzed.

Thawing of frozen scallop meats was performed by placing the scallop meats individually on a wire gauge set on top of a plastic cup. The whole apparatus was enclosed in a plastic bag to prevent evaporation and kept at +4°C in a refrigerator for 12 h. Subsequently, sensory, expressible fluids and instrumental texture determinations were performed on each frozen/thawed scallop meat. The remaining tissue from those determinations and a slice of approximate weight of 2 g, which was removed from the right surface of each scallop meat were used for the preparation of the extracts for the chemical and biochemical analyses, as follows (i) the slices from 5 scallop meats were used for the preparation of one extract for the β-hydroxy-acyl-coenzyme-A dehydrogenase (HADH) determinations and (ii) the remaining tissues from 5 scallop meats were pooled first and then minced in a domestic mincer. The minced scallop meats were then immediately used for the preparation of extracts.

For the chemical and biochemical analyses, 3 extracts were prepared for each storage period coming from 5 different scallop meats. The extracts were stored in an -80°C freezer until analysis.

The results of the chemical and biochemical analyses were the mean of 3 independent determinations and those of sensory, expressible fluids and instrumental texture determinations were the mean of 15 independent determinations.

Freezing time and rate determination

The thermocouples for the temperature measurements were placed at the centre of the thickest part of the scallop meats which was taken as the maximum distance between the right and the left surface of the scallop meats, and was measured by using a vernier instrument. The thermocouple was inserted at the centre of the thickest part from the lateral surface of the scallop meat.

The freezing time (t_e) was calculated as the time (minutes) required to decrease the temperature of the thermal centre from an average initial temperature of 4 ± 1°C to a final temperature of -20°C following the recommendations of the International Institute of Refrigeration (1986). The freezing rates at the thermal centre, expressed as cm h^{-1}, were obtained from the ratios of the distance from the surface to the thermal centre of the scallop muscles and the effective freezing times, t_e (in hours; Chen and Pan, 1995) following the formula

Freezing rate (cm h^{-1}) = Half thickness of scallop muscle (cm) x t_e^{-1}

The characteristic freezing time (t_c) was calculated according to Bevilaqua et al. (1979) as the time (in min) for which the thermal centre of scallop muscles was in the temperature range of maximal ice crystallization, that is from -1 to -7°C.

Determination of the β-hydroxy-acyl-coenzyme-A dehydrogenase activity of scallop meats

The filtrates for the HADH (enzyme class [EC] 1.1.35) assays were prepared according to Fernandez et al. (1999). The HADH released in the filtrate was assayed according to Fernandez et al. (1999). Results were expressed as milliunits per gram of tissue.

Ca^{2+}- ATPase activities in actomyosin extracts

For the preparation of actomyosin, a portion (5 g) of the scallop meats' mince was washed with 25 ml of ice-cold de-ionised water for 15 min and drained through a chilled Buchner No 3 funnel under vacuum. This step was needed to deplete the mince of sarcoplasmic proteins. It was repeated twice more.

A volume of 20 ml of iced-cold 5% (w/v) NaCl (pH = 7) was added to the washed mince. The slurry was allowed to stand at 0 to 4°C for an extraction period of one hour and subsequently was centrifuged for 30 min at 5,000 g at 4°C. The supernatant solution was designated 'actomyosin extract' and was used for protein content and Ca^{2+}-ATPase activity measurements.

The protein concentration in actomyosin extracts was determined by the bicinchoninic acid (BCA) procedure (Sigma Procedure TPRO-562, BCA-1, Sigma Biochemicals Co., St. Louis, Mo., U.S.A).

The Ca^{2+}-ATPase activity was determined according to Carvajal et al. (1999). A portion (100 µl) of actomyosin extract was added to 50 µl of 0.5 M Tris-maleate (pH = 7). To that mixture were then added 50 µl of 0.1 M calcium chloride, 750 µl de-ionized water and 50 µml 20 mM ATP solution (pH = 7). The reaction was conducted for exactly three minutes at 25°C and terminated by adding by adding 0.5 ml of chilled 15% (w/v) of trichloroacetic acid solution (TCA). The mixture was then centrifuged at 10,000 g for 30 min and the inorganic phosphorus liberated in the supernatant was measured by the method of Fiske and Subbarow (1925). Specific activity was expressed as µmoles inorganic phosphate (Pi) released/mg protein/minute. A blank solution was prepared by adding chilled TCA prior to addition of ATP.

Thiobarbituric acid reactive substances determinations

A portion (5 g) of the scallop meats' mince was homogenized with 20 ml chilled 7.5% (w/v) trichloroacetic acid (TCA) for 0.5 min and filtered using Whatman No 1 filter paper. A portion (0.5 ml) of this filtrate was used for the determination of thiobarbituric acid reactive substances (TBARS, mg malondialdehhyde per kg of sample) according to Vyncke (1970).

Determinations of expressible fluids

Two cylindrical portions of each scallop meat, 4 mm in thickness and 20 mm in diameter, were excised from the left surface of scallop meats by means of a ring having 4 mm thickness and 20 mm diameter. Each cylinder was weighed accurately with a Mettler analytical balance and placed on a double thickness filter paper Whatman No 1. It was then covered with another double thickness filter paper and the pack was subjected to a 1,000 g force for 1 min using a Steven's texture analyzer fitted with a cylindrical flat probe (50 mm diameter and 20 mm height). The above-mentioned force was chosen since it would cause the least possible damage to the cylinders (Chung and Merritt, 1991a). Expressible fluids were calculated from the weight difference between the initial and the final weight of the cylinders. Two measurements per frozen stored and thawed scallop meat were taken.

The results of expressible fluids determinations were expressed as g per kg of weight of frozen/thawed scallop meats.

Texture determination as measured by the texture analyzing system

Texture measurements were performed according to Chung and Merritt, (1991b). Peak shear force measurements were performed with a Steven's texture analyzer at a crosshead speed of 50 mm per min. Shear strengths were measured with a V-shaped Warner-Bratzler shear probe mounted on the Steven's load cell.

From the central part of each scallop meat 2 cylinder portions (10 mm in diameter and 10 to 15 mm long) were excised longitudinal to muscle fibres using a cork borer. Individual cylinders were weighed and then inserted through the triangular opening of the blade and placed on the load cell in such a position that the scallop meat fibres were at right angles to the blade penetration. The peak shear force, expressed in gram-force (g*), required to cut the cylinder into 2 pieces was read from the control panel of the analyzer. The shear peak force was adjusted to units of g*per g of scallop meat cylinder to take into account of variations in weight of scallop meats cylinders (Chung and Merritt, 1991b).

Sensory assessments

A portion (4-5 g) of muscle was removed from the anterior part of each scallop meat longitudinal to muscle fibres. It was steam-cooked for 10 min using a domestic steam cooker. After cooking, the portions of scallop meats were placed on wire gauze supported on a plastic cup, the whole apparatus was covered with a polyethylene bag and left at room temperature to cool for 30 min. The portions of scallop meats were then evaluated by 15 trained assessors for flavour, texture and overall acceptability.

Flavour and overall acceptability were scored on a 5-point scale and texture on a 6-point scale. The scoring system for flavour was as follow:

5 = normal sweet taste
4 = no sweet flavour, neutral
3 = slight sour or rancid flavour
2 = moderately strong sour and rancid flavour
1 = very unpleasant putrid or rancid flavour

The degree of liking (overall acceptability) was rated using a five point hedonic scale. The scale was as follows:

5 = like very much
4 = like
3 = neither like or dislike
2 = dislike
1 = dislike very much.

For texture rating, the scoring system, described by Koning and Mol (1991) with slight modifications, was used. The scoring system was as follow:

5 =Tender, succulent, normal texture
4 = Slightly tough and dry
3 = Tough, dry but edible

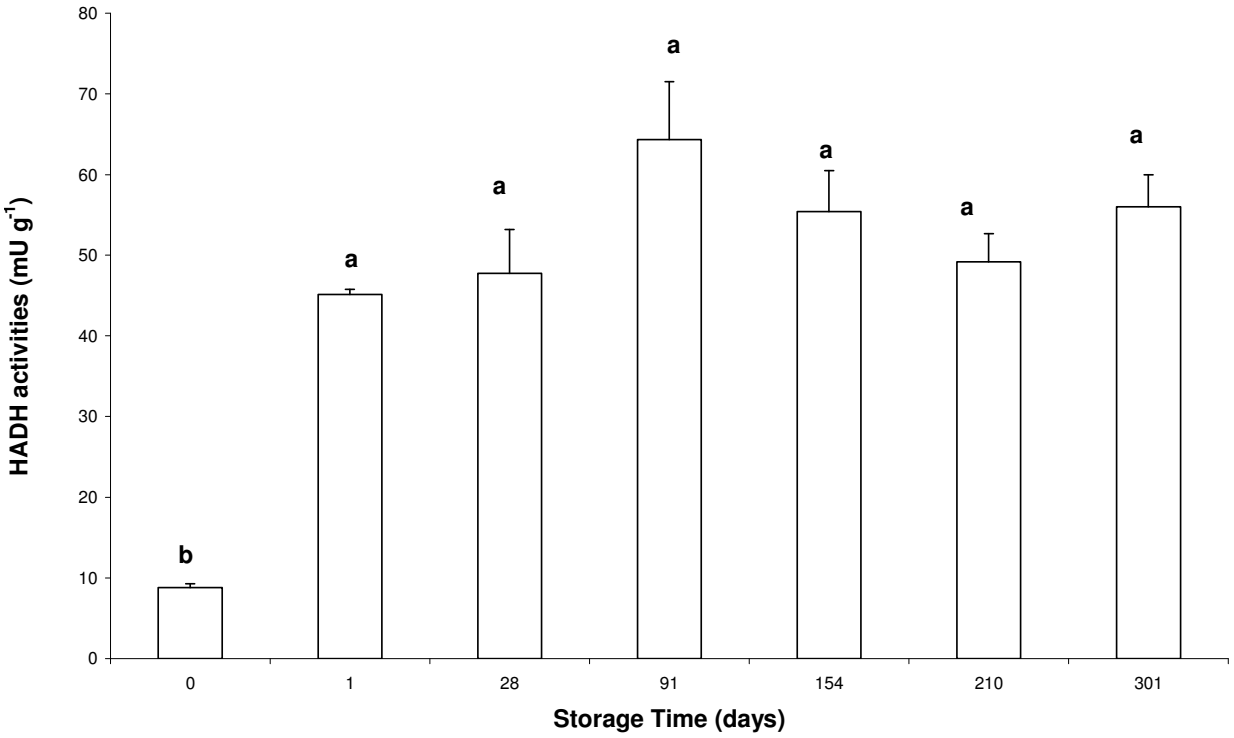

Figure 1. The effect of the length of time of storage at -22˚C on β-hydroxy-acyl-coenzyme-A dehydrogenase activities. Values are means ± SEM, n = 3. Groups with different letters (a, b) are significantly different (P<0.05). The '0' storage time presents fresh scallop meats.

2 = Very tough and dry
1 = Stringy, unable to swallow
0 = Very stringy, completely inedible.

Statistical analyses

One-way analysis of variance (ANOVA) was performed to test for the effects of storage time on physical, chemical and biochemical parameters. ANOVA based on Kruskal-Wallis method was used to examine the effects of storage time on each sensory attribute, since scoring was limited to discrete values. Pearson's correlation coefficients between the means of the parameters studied were also calculated. The parametric and non parametric ANOVAs showing significant differences were followed by a Tukey HSD and Dunn post-hock test, respectively. In all statistical analyses significance was accepted when $P < 0.05$ (Zar, 1984).

RESULTS AND DISCUSSION

The freezing process

Freezing scallop meats in a -80°C freezer produced freezing times, freezing rates and characteristic freezing times equal to 28 min, 3.17 cm/h and 19 min, respectively. In commercial fish industries, scallop meats are frozen mostly individually in mechanical freezing systems (Mason, 1983; Hardy and Smith, 1986). Typical freezing rates that can be met by freezing of seafood by such freezing systems range from 0.3 to 3 cm/h (International Institute of Refrigeration, 1986). Therefore, the experimental conditions of freezing scallop meats in the present study produced freezing times and rates that can be met in commercial practice of freezing scallop meats.

Effect of storage time on β-hydroxy-acyl-coenzyme-A dehydrogenase activity in scallop meats

In fresh tissue the enzyme HADH is retained in mitochondria. However, HADH activities were found in the filtrates of fresh scallop meats (Figure 1). These results suggest that mitochondria were damaged at the surfaces where the scallop muscles were cut and thus a certain amount of the enzyme had leaked from damaged mitochondria into the muscle (Gottesman and Hamm, 1983). Moreover, autolysis of the scallop meats could have caused disruption of some mitochondria, since the fresh scallop meats were stored in ice for 3 days prior to analysis (Hoz et al., 1992, 1993; Pavlov et al., 1994).

The activities of HADH in scallop meats frozen for 24 h at -80°C and then thawed were significantly higher than those in fresh scallop meats (Figure 1). HADH activities have been reported to have increased due to freezing in trout (Garcia de Fernando et al., 1992; Hoz et al., 1992), kuruma prawn (Hoz et al., 1993), sole, salmon, prawn, Norwegian lobster (Fernandez et al., 1999), plaice, whiting and mackerel (Dulfos et al., 2002). These findings

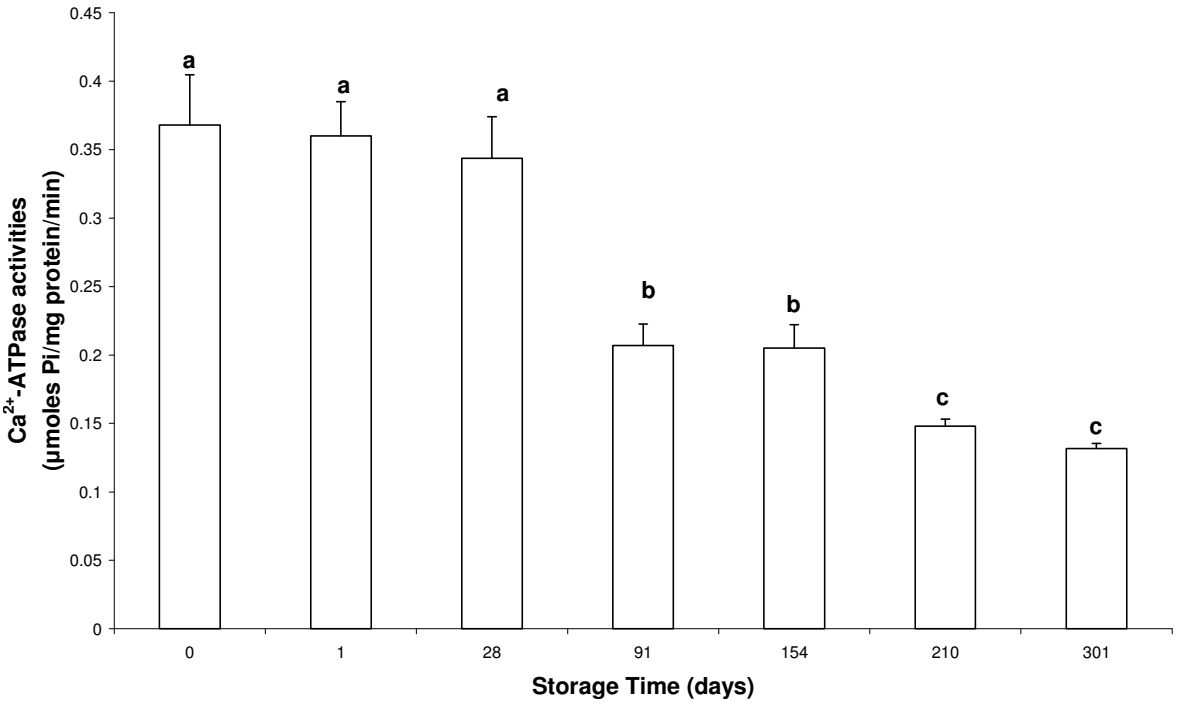

Figure 2. The effect of the length of time of storage AT -22˚C on Ca^{2+}-ATPase activities. Values are means ± SEM, n = 3. Groups with different letters (a, b, c) are significantly different (P<0.05). The '0' storage time presents fresh scallop meats.

and the results of the present study indicate that the freeze-thaw process itself affects the integrity of mitochondria of scallop meats. The freeze damage of mitochondria may be due to dehydration of the mitochondrial inner membrane and/or mechanical disruption of mitochondrial membranes by ice crystals (Hamm and Gottesmann, 1982).

From the results of the present study, an increase in HADH activity in scallop meats was observed in the first 91 days of storage at -22℃, followed by a slight decrease up to 301 days (Figure 1). HADH activities in frozen scallop meats stored frozen for 91 days were 7.3 times the HADH activities of fresh scallop meats. In addition, the HADH activities in the scallop meats, frozen for 24 h at -80℃ and then thawed increased about 4.5 times the activity of the fresh scallop meats. Thus, a part of the release of HADH in frozen scallop meats, stored for 91 days at -22℃, may be related to the time the scallop muscles remained in frozen storage at -22℃ and not only to the freezing process. By means of lysosomal marker enzymes, Benjakul et al. (2003) showed that storage over 24 weeks at -18℃ caused disintegration of membrane structures of several tropical fish.

The storage of a frozen muscle at temperatures above its eutectic temperature goes together with the growth of intra-cellular ice and formation and accretion of inter-cellular ice (Hamm, 1986). In the present study, scallop meats were frozen at characteristic freezing time (t_c value) of 19 min, which should, mainly, result in intra-cellular ice formation (Love, 1955). Therefore, the additional release of the HADH enzyme, observed in scallop meats stored for 91 days at -22℃, may be due to further mechanical damage of mitochondrial membranes by the enlargement of intra-cellular ice and/or dehydration of membranes by the formation and accretion of inter-cellular ice (Hamm and Gottesmann, 1982). Moreover, the decrease in HADH activity in scallop meats after 91 days of storage may be due to the denaturation of the released enzyme during prolonged storage. Similar results and suggestions were reported by Benjakul et al. (2003) after studying the effects of prolonged cold storage at -18℃ on membrane integrity of croaker and lizardfish as determined by marker lysosomal enzymes.

Overall, the time in cold storage at -22℃ may affect the integrity of intra-cellular organelles of scallop meats via ice recrystallization, it may also affect the activity of released mitochondrial enzymes.

Effects of storage time on Ca $^{2+}$-ATPase activities in actomyosin extracts

Ca^{2+}-ATPaseactivity in actomyosin extracts from frozen stored frozen scallop meats decreased throughout the 301 days of frozen storage at -22℃. The marked decrease was observed within the first 91 days of frozen storage (Figure 2).

The decrease in Ca^{2+}-ATPase activities in actomyosin

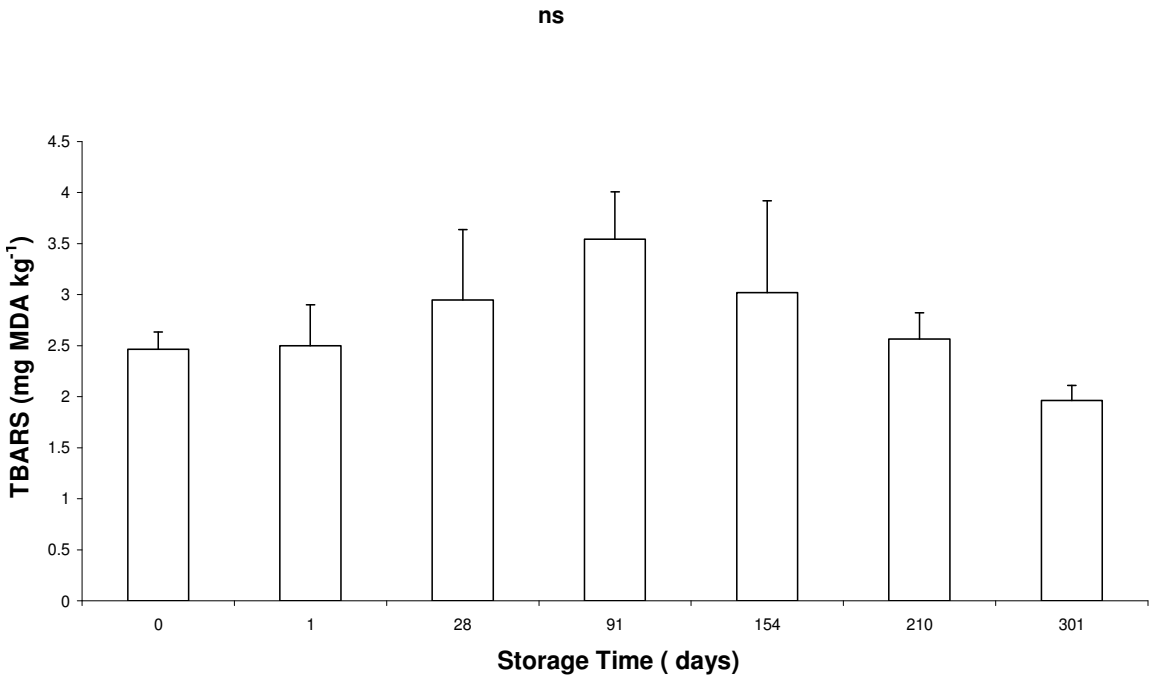

Figure 3. The effect of the length of time of storage at -22˚C on thiobarbituric acid reactive substances (TBARS). Values are means ± SEM, n = 3, ns = not significant. The '0' storage time presents fresh scallop meats

extracts during extended frozen storage of scallop meats could indicate conformational changes (unfolding) and aggregation of the head region of myosin (or 'acto-myosin'), which contains the active site of the enzyme. Re-arrangements of protein via protein -protein interactions might have contributed also to the loss in AT Pase activity. These changes in myosin (or 'actomyosin') might have been caused by the increased salt concentration in the unfrozen phase of scallop meats as a consequence of ice re-crystallization. Similar suggestions were reported by Benjakul et al. (2005) after studding the effects of frozen storage on Ca^{2+}-ATPaseactivity in actomyosins extracted from several tropical fish at -18°C for up to 6 months.

Decrease in Ca^{2+}-ATPaseactivities in mackerel and amberfish muscle was observed during storage at -10 and -40°C for up to 6 months (Jiang et al., 1985). Ca^{2+}-ATPase in Alaska Pollock decreased during frozen storage at -29 °C for 9 months (Scott et al., 1988). Ca^{2+}-ATPase in myctophid species decreased during frozen storage and the degree of decrease depended on the fish species (Seo et al., 1997). Ca^{2+}-ATPase activities decreased in croaker, lizardfish, threadfish bream and bigeye snapper during storage at -18°C for 24 weeks (Benjakul et al., 2003). Tejada et al. (2003) showed that the Ca^{2+}-ATPase activities decreased in the muscles of whole frozen stored gilthead seabream and hake after one year at -20°C.

The results of the present study with stored frozen scallop meats are, therefore, in agreement with these other studies.

Effects of storage time on thiobarbituric acid reactive substances (TBARS)

From the results of the present study, TBARS did not change significantly during storage of frozen scallop meats at -22°C. However, TBARS reached a maximum up to 91 days of storage and then decreased (Figure 3). Changes in TBARS during cold storage have been studied in chub mackerel and smooth hound (Vareltzis et al., 1988), horse mackerel and hake (Simeonidou et al., 1997), with blue whiting and light and dark muscles of hake (Aubourg, 1999; Aubourg et al., 1999), albacore tuna (Ben-Gigirery et al., 1999) and Nile perch (Namulema et al., 1999). In most of these cases TBARS reached a maximum and then fluctuated or decreased. TBARS is a measure of malondialdehyde, which is an end-product of lipid oxidation. The decrease of malondialdehyde in frozen stored fish muscle was attributed to interactions of malondialdehyde with amines, nucleosides, nucleic acids, aminocontaining phospholipids, proteins or other by-products of lipid oxidation (Aubourg et al., 1999).

Effects of storage time on expressible fluids

Expressible fluids from stored frozen scallop meats showed a pronounced increase during the first 91 days of storage at -22°C. From that point until 301 days of storage, there were no significant changes in thawing and total weight losses of frozen stored scallop meats (Figure 4). Several investigations have shown that the quantities

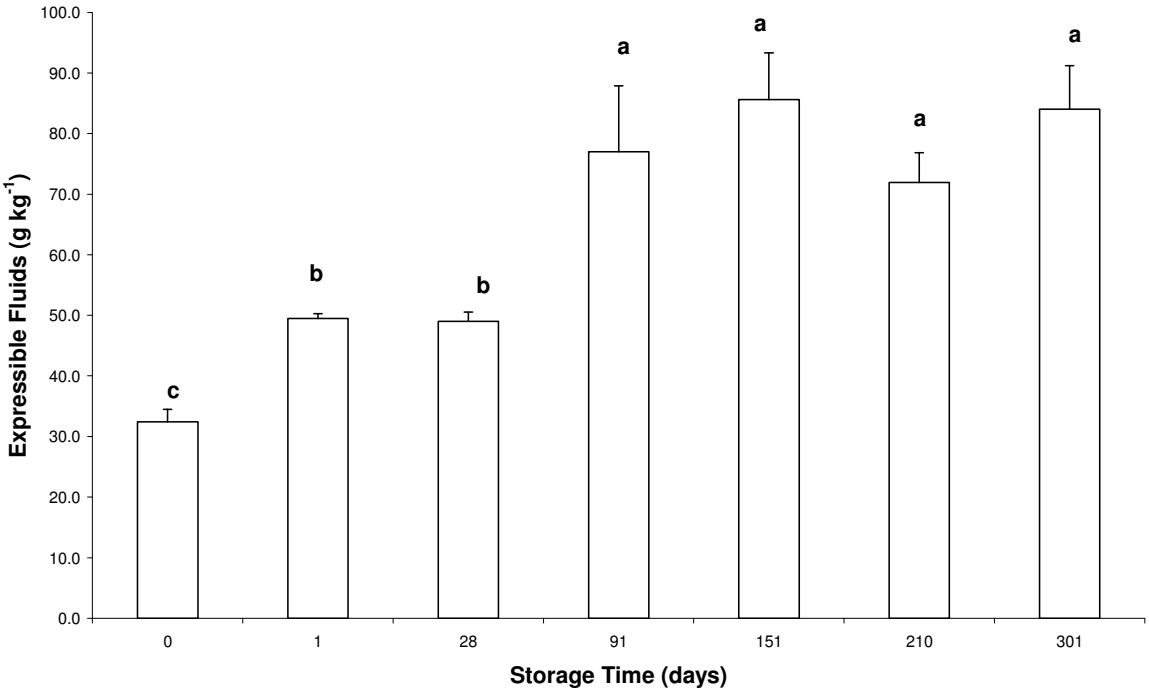

Figure 4. The effect of the length of time of storage at -22˚C on expressible fluids. Values are means ± SEM, n = 15. Groups with different letters (a, b, c) are significantly different (P<0.05). The '0' storage time presents fresh scallop meats

of exudates are influenced from the time a meat product is kept in the frozen state. This was the case with frozen stored turbot (Chevalier et al., 2001), round sardines (Suarez et al., 2002), whole and gutted seabream (Huidobro and Tejada, 2004) and Nile perch (Namulema et al., 1999; Natseba et al., 2005).

Therefore the length of time of storage at -22°C affected the quantities of the exudates of frozen scallop meats.

Expressible fluids were related significantly with Ca^{2+}-ATPase activities in actomyosin extracts from frozen stored scallop meats (Table 1). This observation suggests that the water holding capacity of frozen stored scallop meats was possibly affected by the denaturation of myofibrillar proteins, as measured by the Ca^{2+}-ATPase activities in actomyosin extracts. Similar suggestions were reported by Benjakul et al. (2003) for several tropical fish species.

Effects of storage time on instrumental texture Measurements

The peak shear forces obtained from the fresh scallop meats were significantly higher than those of frozen and immediately thawed scallop meats (Figure 5). This means that the freezing process itself caused softening of raw scallop meats. Freezing and thawing cause lysis of lysosomes and release into sarcoplasm of proteases, which cause breakdown of muscle proteins and consequent tissue-softening (Civera et al., 1996; Pan and Yeh, 1993). Therefore, it is likely that the freezing process itself causes release of proteolytic enzymes from lysosomes of scallop meats with concomitant softening of their tissue.

From the results of the present study, there was no sign of a tendency for peak shear values of frozen scallop meats to change in up to 301 days of storage at -22°C (Figure 5). Similarly, it has been reported that shear strength values from frozen stored gilthead seabream did not change in up to 9 months of storage at -20°C (Pastor et al., 1999). Tissue toughening is common to many low-fat fish species stored at subzero temperatures. This is the case with minced European hake (Careche and Tejada, 1991), cod and haddock (Badii and Howell, 2002). The changes in the texture of fish muscle during frozen storage have been associated with the denaturation and aggregation of myofibrillar proteins, among other causes (Haard, 1992; Mackie, 1993). The composition of scallop meats is somewhat similar to white flesh of less fatty fish (Webb et al., 1969). Moreover, the results of the present study show a significant effect of storage time on denaturation of actomyosin extracted from frozen stored scallop meats. Thus, scallop meats were expected to toughen with time in frozen storage. However, factors related to tissue softening, e.g. release of proteases from lysosomes and cell disruption by ice crystals formation, may also exist in frozen and thawed seafoods and counteract the tissue-toughening factors

Table 1. Pearson's correlation coefficients between parameters of the stored frozen scallop meats.

	Storage time	HADH	Expressible Fluids	Ca^{2+}-ATPase	TBARS	Instrumental Texture
Storage Time	1					
HADH	0.343	1				
Expressible Fluids	0.782	0.697	1			
Ca^{2+}-ATPase	-0.929**	-0.561	-0.860*	1		
TBARS	-0.527	0.470	-0.043	0.219	1	
Instrumental Texture	0.083	0.376	-0.023	-0.312	0.455	1
Flavour	-0.938**	-0.481	-0.819*	0.921*	0.472	-0.040
Texture	-0.987**	-0.443	-0.799*	0.964**	0.447	-0.182
Acceptability	-0.953**	-0.442	-0.851*	0.964*	0.412	-0.091

*Significant at level 5%. ** Significant at level 1%. Numbers without asterisk are not significant; degree of freedom = 5. Numbers in bold imply a strong relationship between the parameters.

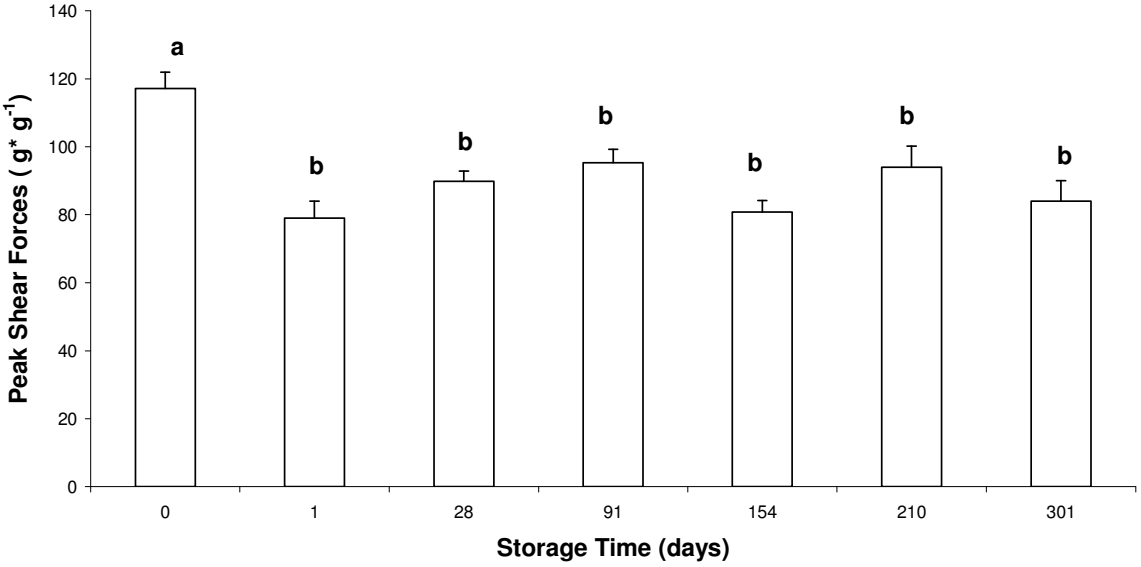

Figure 5. The effect of the length of time of storage at -22˚C on peak shear forces. Values are means ± SEM, n = 15. Groups with different letters (a, b) are significantly different (P < 0.05). The '0'storage time presents fresh scallop meats.

(Srinivasan et al., 1997).

Thus, it is likely that factors which caused the two opposing effects, i.e. tissue-softening and tissue-toughening, might both have been active in frozen stored scallop meats; because of their counteraction, individual effects on textural changes in stored frozen scallop meats up to 301 days at -22°C might have been diminished.

Effects of storage time on sensory attributes

According to the ratings of the sensory panel, the fresh scallop meats had a sweet to neutral taste and tender to slightly tough texture (ratings between 5 and 4). The frozen scallop meats stored for 301 days had neutral to slightly sour and rancid taste and slightly tough to tough and dry texture (ratings between 4 and 3; Table 2).

The overall acceptability ratings of frozen scallop meats stored at -22°C decreased significantly by 210 days (Table 2), but the mean acceptability score was more than 3 (that is, 'neither like or dislike') after 301 days of storage indicating that the scallop meats remained acceptable at the end of the storage period. Changes in flavour, texture and acceptance of stored frozen seafoods have been recorded by other workers. These changes are mainly dependant on species, pre-freezing treatment and time and temperature of frozen storage. Thus, Simeonidou et al. (1997) found that the taste and texture of horse mackerel and Mediterranean hake were reduced during 360 days of storage at -18°C, but these attributes

Table 2. The effect of the length of time of storage at -22˚C on sensory attributes of scallop meats*.

Storage time (days)	Flavour	Texture	Acceptability
0	4.6 ± 0.11 a	4.6 ± 0.14 a	4.2 ± 0.10 a
1	4.5 ± 0.15 a	4.6 ± 0.22 a	4.2 ± 0.20 a
28	4.8 ± 0.13 ab	4.6 ± 0.20 ab	4.4 ± 0.20 a
91	4.2 ± 0.20 ab	4.1± 0.17 ab	3.8 ± 0.20ab
154	4.17 ± 0.17 ab	4.0 ±0.19 ab	3.6 ± 0.30 ab
210	4.0 ± 0.25 ab	3.8 ±0.23 ab	3.3 ± 0.14 b
301	3.6 ± 0.15 b	3.5 ± 0.22 b	3.1 ± 0.20 b

*Means ± S.E.M, n = 15. Values in the same column with different letter (a, b) are significantly different ($P < 0.05$). The '0' storage time presents fresh scallop meats.

were still at an acceptable level at the end of the storage period. Namulema et al. (1999) showed that the texture, taste and overall acceptability of stored frozen Nile perch at -27°C for 10 weeks were similar to fresh fish. Yilmaz and Akpinar (2003) showed, also, that the frozen stored fillets of guitarfish at -18°C were acceptable after 6 months of storage. In addition, post-rigor scallop meats, frozen within 6 days after shucking, were acceptable after 6 months at -30°C (Chung and Merritt, 1991a).

The results of the present study are in agreement with these other studies and suggest that there was a loss in flavour, texture and overall acceptability of stored frozen scallop muscles during 301 days storage at -22°C, but that these products were in acceptable condition at the end of the storage period.

Correlations between parameters studied

Ca^{2+}-ATPase activities in actomyosin extracts from stored frozen scallop meats showed a significant correlation with the storage time and sensory attributes, including texture (Table 1). Significant correlations between parameters related to changes in myofibrillar proteins and sensory texture are recorded in the literature for stored frozen fish products. This is the case with stored frozen Patagonian hake at -20 and -30°C, (Ciarlo et al., 1985), stored frozen fillets and minces of hake at -18°C (Koning and Mol, 1991) and stored frozen minced sardines at -18°C (Verma et al., 1995). The commercial significance of these results is that non-sensory parameters which show satisfactory linear correlations with storage time and sensory attributes may be useful methods for assessing the quality of stored frozen seafoods for commercial purposes, which requires customer satisfaction. Therefore, Ca^{2+}-ATPase activities may be reliable method for Industry to use for assessing the quality of frozen scallop meats during long term storage at -22°C.

Conclusions

The results of the present study indicate that the length of time of storage at -22°C affected the integrity of intracellular (mitochondria) organelles, reduced the water holding capacity, caused denaturation of myosin (or 'actomyosin') and affected the sensory attributes (flavour, texture and acceptability) of the frozen scallop meats. Most of these changes in scallop meats were more pronounced after 91 days of storage at -22°C. It can, therefore, be concluded that although the frozen scallop meats were in acceptable condition up to 301 days (that is, almost 10 months), holding of these products for up to three months (i.e. 91 days) at -22 °C may prevent the negative changes in muscle structure, water holding capacity, myofibrillar proteins and sensory quality which occur with longer storage.

Among the different indices checked, Ca^{2+}-ATPase activities in actomyosin extracts may be useful for assessing the quality loss of the scallop meats stored frozen at -22°C, since a good linear correlation was obtained with time and scores of sensory attributes.

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