

Fatty acids profiles in filets *Pampus argenteus* and *Sparidentex hasta* during frozen storage

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Abstract

In this study changes in fatty acids profile during frozen storage at -18°C for (*Pampus argenteus*) and (*Sparidentex hasta*), caught from the Persian Gulf (Bandar Deylam) were studied. Changes in saturated fatty acids (SFAs), monounsaturated fatty acids (MUFAs), polyunsaturated fatty acids (PUFAs), EPA+DHA/C16, n-3/n-6 and high unsaturated fatty acids (HUFA) were investigated during 95 days storage at -18°C. Twenty fatty acids were found in (*Pampus argenteus*) with high percentage of saturated fatty acids (44.06%), polyunsaturated fatty acids (32.17%), high unsaturated fatty acids (27.8%) and monounsaturated fatty acids (22.62%) and for (*Sparidentex hasta*) found high percentage of PUFA(41.39%), HUFA (37.35%), SFA (36.42%) and MUFA (21.76%). The percentage of Fatty acids of SFA and PUFA in fish *Sparidentex Hasta* during this study did not show a significant difference. The percentage of MUFA fatty acids and HUFA during the freezing period were varied so that the maximum amounts of MUFA and HUFA were found on 65 and 0 day respectively. The fish *Pampus Argenteus* were found also the highest percentage of fatty acids of SFA, PUFA and HUFA during freezing on the days 65 and 35, but for MUFA no significant difference during freezing. Percentage of palmitic acid were found (28.06% and 23.72%), stearic acid (7.84% and 10.21%) in Silver pomfret and Progies respectively. The major unsaturated fatty acids of Silver pomfret and Progies were determined as DHA (19.87% and 22.57%), oleic acid (15.02% and 14.55%) respectively. As a result of the frozen storage (up to 95 days), fatty acid groups such as polyunsaturated, high unsaturated and n-3 polyunsaturated, as well as in the n-3/n-6 ratio were decreased and it means that the nutritional value of Silver pomfret and Progies has decreased.

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Introduction

Although freezing is an effective method of food preservation, but the presence of unsaturated fatty acids in fish tissue makes fish products highly susceptible to lipid oxidation and loss in quality (Hultin, 1992). Freezing of fish widely and effectively are used for thousands of years due to high product quality (Persson and Londahl, 1993). Published papers show that the consumption of fish oil containing PUFA prevents and/or cures arterial hypertension (Millar and Waal-Manning, 1992), colon and prostate cancer (Marchioli, 2001, 2002), human breast cancer growth (Rose and Connoll, 1993), inflammatory diseases (Belluzi *et al.*, 1993; James and Cleland, 1996), asthma (Dry and Vincent, 1991; Hodge *et al.*, 1996), and disorders of the immune system (Levine and Labuza, 1990). Besides, eicosapentaenoic acid (EPA, C20:5n3) and docosahexaenoic acid (DHA, C22:6n3), found

only in fish and sea foods, play a vital role in the development and functioning of the nervous system (brain), photoreception (vision), and the reproductive system (Alasalvar *et al.*, 2002; Skonberg and Perkins, 2002; Tapiero *et al.*, 2002; Sidhu, 2003). It is generally recognized that PUFA and HUFA composition may vary among fish species. Degradation of PUFA and HUFA by auto oxidation during storage and the processing of fish oils and fatty fish easily lead to the formation of volatiles associated with rancidity (Pazos *et al.*, 2005) and therefore, it is both lipid and PUFA and HUFA contents of fish that play a deciding role in its health benefits. Although PUFA and HUFA contents constitutes varied amounts body constitutes among fish species, little attention has been paid to and little information obtained regarding changes in fatty acids in different species during the frozen storage process and period. Fish is one of the best sources of animal protein available which has been widely accepted as a good source of protein and

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Table 1. Changes of fatty acids profile *Pampus argenteus* to 95 days freezing in -18°C

Fatty acid	Day 0	Day 35	Day 65	Day 95
C14:0	5.58±0.52 ^a	3.98±0.43 ^a	5.68±0.61 ^a	2.96±0.36 ^a
C16:0	28.06±0.91 ^b	28.13±0.51 ^b	29.14±1.53 ^b	25.28±0.8 ^b
C16:1	3.10±0.13 ^{bc}	2.78±0.37 ^b	3.57±0.25 ^b	5.52±0.68 ^b
C17:0	0.53±0.05 ^d	1.08±0.07 ^b	1.54±0.12 ^a	0.09±0.06 ^a
C17:1	0.53±0.12 ^b	0.44±0.03 ^b	1.23±0.16 ^b	0.65±0.1 ^b
C18:0	7.34±0.26 ^a	3.54±0.32 ^{ab}	9.06±0.6 ^a	12.4±0.23 ^a
C18:1(n-7)c	15.02±0.58 ^a	14.8±0.37 ^a	18.52±2.13 ^b	0.48±1.65 ^b
C18:1(n-7)t	2.8±0.38 ^b	2.73±0.31 ^b	3.13±0.03 ^b	19.98±0.03 ^a
C18:2(n-6)	2.14±0.32 ^a	2.24±0.27 ^a	2.82±1.79 ^a	1.02±0.05 ^a
C18:3(n-3)	0.33±0.12 ^a	0.4±0.2 ^a	0.46±0.04 ^a	0.914±0.01 ^a
C20:0	0.63±0.06 ^b	0.41±0.11 ^a	0.85±0.05 ^a	0.314±0.01 ^a
C20:3	0.77±0.03 ^a	0.26±0.1 ^a	0.3±0.04 ^a	0.58±0.03 ^a
C20:3	1.57±0.16 ^b	0±0.0 ^a	0.06±0.01 ^a	2.46±0.14 ^b
C20:4(n-6)	1.84±0.27 ^b	2.74±0.35 ^b	1.89±0.21 ^b	2.28±0.17 ^b
C22:1	1.06±0.01 ^a	0.55±0.1 ^b	0.42±0.05 ^b	0.3±0.01 ^b
C20:5(n-3)EPA	3.63±0.15 ^b	3.55±0.19 ^b	2.37±0.64 ^b	2.56±0.34 ^{bc}
C22:4	0.3±0.07 ^a	0.5±0.03 ^a	0.58±0.11 ^a	1.9±0.07 ^b
C22:5	1.14±0.11 ^b	1.91±0.15 ^a	1.02±0.02 ^b	0.45±0.12 ^a
C22:5	1.71±0.07 ^a	1.84±0.1 ^a	2.02±0.12 ^a	4.6±0.16 ^b
C22:6(n-3)DHA	19.87±1.73 ^b	23.02±1.92 ^a	15.03±1.21 ^a	15.26±1.62 ^b
Total SFA	44.06±1.68 ^{ab}	42.16±1.15 ^a	46.28±1.78 ^b	41.04±1.36 ^a
Total MUFA	22.62±0.97 ^a	21.32±0.97 ^a	25.91±0.42 ^b	26.93±0.69 ^b
Total PUFA	32.17±1.82 ^b	35.74±2.12 ^c	25.71±0.7 ^a	29.54±1.64 ^b
Total HUFA	27.8±2.03 ^b	31.97±1.97 ^c	22.59±1.01 ^a	22.03±1.03 ^b
Total n3	27.13±1.77 ^b	28.83±1.84 ^b	19.97±1.25 ^a	25.78±1.45 ^b
Total n6	3.24±0.24 ^a	7.41±0.39 ^b	6.32±1.46 ^b	5.65±0.78 ^b
DHA/EPA	5.96±0.23 ^b	6.74±2.41 ^b	6.47±0.32 ^b	5.47±0.33 ^b
n3/n6	3.37±0.45 ^a	3.88±0.19 ^{ab}	3.28±0.82 ^a	4.56±0.38 ^b

Data are means of experiment examined in duplicate

Means in the same row with different letters differ significantly at $P \leq 0.05$

other elements for the maintenance of healthy body (Arannilewa *et al.*, 2005). Marine lipids have a high content of polyunsaturated fatty acids (PUFAs), particularly eicosapentaenoic (EPA; 20:5x-3) and docosahexaenoic acids (DHA; 22:6x-3) (Pazos *et al.*, 2005; Bayir *et al.*, 2006). There is strong evidence that consumption of fish is favorable to people health (Bayir *et al.*, 2006). Freezing and frozen storage have largely been employed to retain fish sensory and nutritional properties (Lugasi *et al.*, 2007). Silver pomfret fish with scientific names (*Pampus argenteus*) belongs to the family Stromatidae as Bank of migratory fish and the economic value of commercial fish in coastal waters around the Persian Gulf and Sea of Oman. Progies with the scientific name (*Sparidentex hasta*) native Persian Gulf, Indian Ocean and West coast of India. This fish as a high value for the reproduction and breeding of native species. According research results of Javaheri baboli *et al.*, (2012) that fatty acid profile white leg shrimp west after 3 months of frozen storage was considerable variation so that the percentage of SFA, MUFA, PUFA, n3 and n6 fatty acids was affected the 3-months period that showed significant differences. Therefore, according to the commercial

and nutritional value of fish *Pampus argenteus* and *Sparidentex hasta* we decided to study the impact of freezing on the two above-mentioned fish species during the three months storage at -18°C .

There is no information of the fatty acids of *Pampus argenteus* and *Sparidentex hasta* of Iran. The aim of this study was to study on the changes in fatty acids composition during frozen storage in -18°C for 95 days and to evaluate their nutritional value to provide scientific data for food processing and pharmaceuticals.

Materials and Methods

Sample collection and preparation

Fresh samples of (*Pampus argenteus*) and (*Sparidentex hasta*) with weight of about 750 ± 15.38 g and 350 ± 20.2 g were purchased from a local market of Behbahan, Iran. They were immediately brought to the lab in polythene bags along with crushed ice. The viscera of fish were removed; the fish was washed with water and cut into pieces. These pieces were washed and immediately wrapped in aluminium foil, kept in air tight plastic container and stored at -18°C (frozen storage) then analyzed on 0, 35, 65 and 95

Table 2. Changes of fatty acids profile *Sparidentex hasta* to 95 days freezing in -18°C

Fatty acid	Day 0	Day 35	Day 65	Day 95
C14:0	1.49±0.14 ^a	2.41±0.97 ^b	2.37±0.16 ^b	2.96±0.12 ^b
C16:0	23.72±0.31 ^a	26.58±9.82 ^c	24.2±0.68 ^{ab}	25.28±0.5 ^{bc}
C16:1	2.99±0.31 ^a	5.97±2.73 ^b	5.64±0.54 ^b	5.32±0.29 ^b
C17:0	0.8±0.1 ^c	0.52±0.72 ^b	0.72±0.06 ^{bc}	0.09±0.03 ^a
C17:1	0.37 ±0.04 ^a	32.34±0 ^a	1.53±0.11 ^c	0.65±0.32 ^a
C18:0	10.21±2.21 ^a	8.57±2.93 ^a	8.3±0.31 ^a	8.7±0.1 ^a
C18:1(n-9)c	14.55±4.55 ^a	16.51±6.11 ^a	15.9±1.53 ^a	15.48±1.67 ^a
C18:1(n-9)t	3.68±0.36 ^a	3.14±1.56 ^a	4.27±0.68 ^a	4.98±0.89 ^a
C18:2(n-6)c	4.67±1.53 ^b	2.03±0.87 ^a	2.41±1.26 ^a	1.82±0.03 ^a
C18:3(n-3) ? 3	0.55±0.28 ^a	0±0.0 ^a	0.25±0.1 ^{ab}	0.28±0.02 ^{ab}
C 20:0	0.2±0.24 ^a	0.04±0.07 ^{ab}	0.54±0.25 ^c	0.38±0.05 ^{bc}
C 20:3 ? 9	0.17±0.29 ^a	0.16±0.16 ^a	0.34±0.06 ^a	0.25±0.18 ^a
C20:3 ? 3	0.34±0.43 ^a	0.11±0.06 ^a	0.13±0.04 ^a	1.46±0.32 ^a
C20:4(n-6)ARA	5.29±1.76 ^a	4.35±1.74 ^a	5.06±0.59 ^a	5.28±0.43 ^a
C22:1	0.15±0.25 ^a	0.6±0.46 ^a	0.24±0.03 ^a	0.3±0.01 ^a
C20:5(n-3)EPA	4.27±0.97 ^a	2.79±1.24 ^a	3.8±0.19 ^a	3.56±0.61 ^a
C22:4 ? 6 DTA	0.2±0.02 ^a	0.91±0.4 ^b	1.36±0.12 ^b	1.12±0.08 ^b
C22:5 ? 6	0.17±0.29 ^a	1.15±0.43 ^a	0.15±0.02 ^b	0.2±0.01 ^a
C22:5 ? 3 DPA	3.52±1.21 ^a	2.81±1.25 ^a	3.74±0.25 ^a	3.62±0.23 ^a
C22:6(n-3)DHA	22.57±6.02 ^a	16.8±1.41 ^a	18.86±0.89 ^a	18.26±1.27 ^a
Total SFA	36.42±4.18 ^a	38±0.56 ^a	36.15±0.24 ^a	37.41±1.81 ^a
Total MUFA	21.76±4.21 ^a	27.23±2.29 ^b	27.62±0.63 ^b	26.73±1.74 ^{ab}
Total PUFA	41.39±8.5 ^a	33.58±3.09 ^a	34.43±0.34 ^a	34.48±2.26 ^a
Total HUFA	37.35±8 ^b	28.07±2.5 ^a	30.4±1.64 ^{ab}	29.2±2.8 ^{ab}
Total n3	31.27±7.89 ^a	22.93±2.4 ^a	26.8±1.34 ^a	27.18±1.72 ^a
Total n6	5.66±1.67 ^a	10.4±1.6 ^b	8.99±1.2b	8.42±1.2 ^b
DHA/EPA	5.24±0.24 ^{ab}	4.69±0.25 ^a	4.96±0.06 ^{ab}	5.12±0.1 ^b
n3/n6	5.61±0.4 ^c	2.22±0.17 ^a	3.07±0.33 ^b	3.22±0.2 ^b

Data are means of experiment examined in duplicate

Means in the same row with different letters differ significantly at $P \leq 0.05$

days of storage.

Fatty acid analysis

The lipids were saponified and esterified for the fatty acid analysis according to the method reported by Metcalfe *et al.* (1966). The fatty acid methyl esters (FAMES) were analyzed on a Unicomp model 4600 gas chromatograph (GC) with a flame ionization detector (FID). The esters were separated on a 30 m×0.22 mm i.d. wall-coated open tubular fused-silica capillary column (30 m×0.25 mm×0.22 µm film thickness, BPX70; SGE, Melbourne, Australia) at isothermal temperature of 190°C with helium as the carrier gas (50 psi) being used to separate the fatty acids. A splitless injector (1.2 µL injection) was also used at 240°C and a FID at 250°C during the separation process. The peaks were identified based on their retention times using fatty acid methyl ester standards and all samples run in triplicate. An internal standard method (C15:0) was employed to calculate the fatty acid composition.

Statistical analysis

Data were presented as mean standard deviation (SD) and subjected to analysis of variance (ANOVA). Significant means were compared by one-way procedure tests at ($P < 0.05$).

Results

The fatty acid composition and changes in during storage (0, 35, 65 and 95 days of storage) in (*Pampus argenteus*) and (*Sparidentex hasta*) is summarized in Tables 1 and 2, respectively. Four fatty acids, docosahexaenoic acid (DHA), oleic acid (18:1n-9), palmitic acid (16:0) and stearic acid (18:0) were particularly abundant in the muscle tissue of both species. In both (*Pampus argenteus*) and (*Sparidentex hasta*), unsaturated fatty acid contents were higher than saturated fatty acids (SFA < PUFA + MUFA), whereas during frozen storage unsaturated fatty acids decreased in contrast to the saturated fatty acids. Oleic acid (C18:1n9) was the main fatty acid among the MUFAs in both of two fish species.

The SFAs were the most abundant fatty acids in the tissues of *Pampus argenteus*, which found 44.06% of the total fatty acids. Among SFAs, those occurring in the highest content in during the storage period were palmitic acid (C16:0) and stearic acid (C18:0) and oleic acid which found highest among monounsaturated fatty acid in two species.

It is noticeable that both linoleic (C18:2n-6) and arachidonic acids (C20:4n-6) were predominant in the total n-6 polyunsaturated fatty acids and Eicosapentaenoic acid (C20:5n-3) and

docosahexaenoic acid (C22:6n-3) were the major fatty acids in total n-3 polyunsaturated fatty acids in fillets of *Pampus argenteus* and *Sparidentex hasta*. The PUFA found 32.17 % and 41.39% of the total fatty acids in *Pampus argenteus* and *Sparidentex hasta*. Distribution of fatty acid in *Pampus argenteus* was as SFA> PUFA>MUFA and in *Sparidentex hasta* was as PUFA> SFA>MUFA but unsaturated fatty acids were more than saturated fatty acids (SFA<PUFA+MUFA). A reduction was observed in the percentage of PUFA from 32.17% to 29.54% in *Pampus argenteus* and 41.39% to 34.48% respectively after 95th days storage under frozen conditions ($P>0.05$). DHA and EPA fatty acids are the most important of the fish fatty acids in nutrition. These two fatty acids were decreased dramatically after 95 days of frozen storage.

The fresh two fish species found high concentration of n3 PUFAs including ecosapentaenoic acid (EPA, C20:5n3), docosahexaenoic acid (DHA, C22:6n3) as the major components. The high content of DHA was found in fresh samples of *Sparidentex hasta* (22.57% of the total fatty acids; whereas *Pampus argenteus* showed lower DHA content. The ratio of n-3/n-6 was 8.37 in the fresh samples *Pampus argenteus* and this ratio decreased to 4.56 after 95 days of frozen storage. The ratio of n-3/n-6 was 5.61 in the fresh samples *Sparidentex hasta* and this ratio decreased to 3.22 after 95 days of frozen storage. A significant increase were found in MUFAs levels in *Sparidentex hasta* after 95 days of storage ($P<0.05$), while the content of PUFAs were significantly decreased ($P<0.05$).

Discussion

In fact, compared to other animal fat, fat fish are particularly rich in EPA and DHA derived from algae and plankton (Nichols *et al.*, 1989). It is known that freezing are widely used to keep nutritional and sensory properties of fish (Erickson, 1997). However, fish quality decreases during frozen storage as a result of increasing time and temperature of storage as some authors demonstrate (Sotelo *et al.*, 1995; Aubourg, 1999). According to reported results by Godwin and Prabhu (2006), the high degree of unsaturation in fish oils increase the vulnerability to lipid peroxidation. Furthermore, the degradation of the PUFAs group decreased by auto-oxidation leads to the formation of volatile compounds that are associated with rancidity (Pazos *et al.*, 2005). Obtained results showed that the frozen storage at -18°C for 95 days found a decrease in the omega-3 content in both fish species. In fresh fillet, palmitic (C16:0) and oleic acids (C18:1n-9)

were the major fatty acids among the saturated and monounsaturated fatty acids of both fish species. These results are in agreement with those reported in several fish species (Chaijan *et al.*, 2006; Saldanha *et al.*, 2007; Pirestani *et al.*, 2010).

Twenty fatty acids were found in *Pampus argenteus* with a higher percentage of saturated fatty acids (44.06%), and polyunsaturated fatty acids (32.17%), High unsaturated fatty acids (27.8%), monounsaturated fatty acids (33.72%). The HUFAs and PUFAs decreased from 27.8 to 22.03% and 32.17 to 29.54% respectively. The amounts of the fatty acid fractions in the fresh sample of *Pampus argenteus* were SFA>PUFA>HUFA>MUFA, however, at the end of the storage, these amounts were changed to SFA>PUFA>MUFA>HUFA. Our study results were similar to the findings on Red tilapia fatty acids changes during storage at -20°C for 30 weeks which reported by Ng and Bahurmiz, (2009).

Twenty fatty acids were found in *Sparidentex hasta* with a higher percentage of polyunsaturated fatty acids (41.39%) and high unsaturated fatty acids (37.35%), saturated fatty acids (36.42%), monounsaturated fatty acids (21.76%). The HUFAs and PUFAs decreased from 37.35 to 29.2% and 41.39 to 34.48% respectively. The results showed that unsaturated fatty acid contents were greater than saturated fatty acids in fillets of both two fishes. the unsaturated fatty acids content in fillets *Pampus argenteus* and *Sparidentex hasta* found 54.79%, 63.15% and saturated fatty acids contents of two species found 44.06% and 36.42% respectively. Similar results were reported (Hedayatifard and Moeini, 2007; Sahari *et al.*, 2009 and Pirestani *et al.*, 2010).

Palmitic acid (C16:0) and stearic acid (C18:0) with 28.06% and 7.84%, respectively were the major fatty acids among the SFAs in fillets *Pampus argenteus* during storage. The same results were obtained about Mackerel and Shark (Sahari *et al.*, 2009) and Sturgeon (Hedayatifard and Moeini, 2007). eicosapentaenoic acid (C20:5n-3) and docosahexaenoic acid (C22:6n-3) were the major fatty acids from total n-3 polyunsaturated fatty acids in fillets of *Pampus argenteus* and *Sparidentex hasta*. eicosapentaenoic acid (C20:5n-3) and docosahexaenoic acid (C22:6n-3) also played a major role in the total n-3 polyunsaturated fatty acids. EPA is the most important essential fatty acids of the n-3 series in human diet because it is the precursor to the 3-series ecosanoids (Chen *et al.*, 1995). It has been reported that DHA decreases the concentration of low density lipoprotein cholesterol in plasma (Child *et al.*, 1990). In fresh and frozen samples DHA content

was more than EPA content although both these fatty acids were decreased at the end of frozen storage ($P < 0.05$). Our study results are consistent with those that characterize several fish species (Soriguer *et al.*, 1997; Garcia-Arias *et al.*, 2003). Considering fatty acid series (Tables 1 and 2), showed the frozen storage led to a progressive content increased in MUFA in fillets *Pampus argenteus* and SFA and MUFA in *Sparidentex hasta*, while SFA, PUFA, n-3 in *Pampus argenteus* and HUFA, PUFA, n-3 in *Sparidentex hasta* decreased with increasing the frozen storage time. Pigott and Tucker (1990) stated that the ratio n3/n6 be the best indicator for measuring the nutritional value of different species of fish oil. The increase the ratio of omega 3 to omega-6 in the human diet prevents heart disease by lowering plasma lipids and reduce the risk of cancer (Kinsella *et al.*, 1990). The amounts of omega-3 to omega-6 ratio recommended by nutritionists greater than 1:4 (Valencia *et al.*, 2006). Additionally, a progressive decrease with frozen time could be observed for the n-3/n-6 ratio. Freezing storage is known to be associated with fish lipid oxidation processes that could be explained as a result of the presence of pro-oxidant enzymes in the fish muscle such as lipoxygenases, peroxidases and chemical pro-oxidant molecules named hemoproteins and metal ions (Sikorski and Kolakowski, 2000). The same results were found (Sahari *et al.*, 2009) for Mackerel (*Scoromorus Commerson*) and Shark (*Carcharhinus Dussumieri*). Generally, Fish unsaturated fatty acids, polyunsaturated, in particular omega-3 fatty acids are important among which the EPA or eicosapentaenoic acid (C20: 5n-3) and docosahexaenoic acid or DHA C22: 6n-3) are more popular. Polyunsaturated omega-3 fatty acids such as DHA and EPA have an important role in human health. In this study, the amounts of EPA and DHA in fish *Pampus Argenteus* significantly were reduced during freezing on day 65 compared to the other time periods, but there was no significant difference in *Sparidentex hasta* fish. The \sum n-3 in *Pampus argenteus* and *Sparidentex hasta* were presented as 27.13% and 31.27% of the total fatty acids, most abundant of which was DHA above 19.87% and 22.57%, respectively. The \sum n-6 in *Sparidentex hasta* was presented as 5.66% of the total fatty acids, most abundant of which was linoleic acid (4.67%) and arachidonic acid (5.29%). Significant increase was observed in the percentage \sum n-6 of fillets *Sparidentex hasta* during 95 days ($P < 0.05$). Marine fish are rich in n-3 fatty acids, especially DHA and EPA (Celik *et al.*, 2005). The \sum n-6 in *Pampus argenteus* were presented as 3.24% of the total fatty acids and were mainly linoleic acid (2.14%) and arachidonic acid (1.84%).

It has been reported that the types and amounts of fatty acids in fish tissues vary with the geographic location, size, age, what the fish eat, reproductive status and season (Celik *et al.*, 2005). In our study, the amount of n-3 in two fish species was more than the n-6 compounds. A significant decrease in this ratio n-3/n-6 of *Pampus argenteus* and *Sparidentex hasta* respectively from 8.37 to 4.56 and 5.61 to 3.22 showed that the nutritional value of this fish had declined during frozen storage. Suggested that the n-3/n-6 ratio is a better index in comparing relative nutritional value of fish oils of different species (Turan *et al.*, 2007; Pirestani *et al.*, 2010). The n-3/n-6 ratio of 1:1 is considered to be optimal for nutritional purposes (Turan *et al.*, 2007). The n-3/n-6 ratio in mackerel and shark were reported (Sahari *et al.* 2009), 4.16 and 2.02, respectively and gilthead sea bream was found between 1.6 to 3.6 in different months (Senso *et al.*, 2007). The n3/n6 ratio is a useful criterion for comparing the relative nutritional values of fish oils. It has been suggested that a ratio of 1:1-1:5 would contribute to a healthy human diet (Osman *et al.*, 2001). Decrease in unsaturated fatty acid content, particularly PUFA, and lower n-3/n-6 ratios were also obtained (Pirestani *et al.*, 2010) in different kinds of fresh water fishes (*Caspian kutum*, golden grey mullet, common carp, pike perch and common kilka) belonging to the South Caspian sea during frozen (-24°C) storage. The frozen storage led to important changes in the fatty acids profile in fillets *Pampus argenteus* and *Sparidentex hasta* (Tables 1 and 2). Results showed an increase with time in some saturated fatty acids in *Pampus argenteus* (C18:0) and in C20:4n-6 (arachidonic acid) and of *Sparidentex hasta* (C16:0, C14:0).

Previous research related to fish frozen storage has already shown that unsaturated lipids are likely to be oxidized. Thus, Serdaroglu and Felekoglu (2005) reported that SFA and PUFA increased and decreased, respectively, in minced sardine (*Sardina pilchadus*) muscle when stored at -20°C up to 5 months. A similar behavior was found for both fatty acids group presence in frozen (-30°C) Spanish mackerel (*Scoromorus commersoni*) and fillets white cheek shark (*Carcharhinus dussumieri*) (Nazemroaya *et al.*, 2009). According to this study, shorter storage time is suggested for frozen fish.

Conclusion

The effects of storage time on fatty acids of two fish species were examined in the study. It was found that the storage time (at -18°C) found a significant impact on the storage stability of fish. The observed

changes in SFA, MUFA, PUFA, HUFA, $n3/n6$ reveal that both two fish species were susceptible to significant change during the frozen storage, especially if the storage time be long. In addition, the decrease in unsaturated fatty acids, especially polyunsaturated fatty acids, $n3/n6$ showed that nutritional values of these species have decreased. Increasing SFA and decreasing PUFA concentrations indicated that oxidation is in progress, and this has an important effect on quality. Based on the present study, all fish species can be stored for 95 days in a frozen state with low undesirable changes of fatty acids profile. However, it is suggested that the effects of frozen storage on fatty acids compounds should be further investigated, in a large scale study, preferably as a socioeconomic evaluation and as well as a thorough evaluation for long term frozen storage on the changes in fatty acids profile. In addition, the decrease in unsaturated fatty acids, especially polyunsaturated fatty acids and $n-3/n-6$ showed that the nutritional value of *Pampus argenteus* and *Sparidentex hasta* fish has decreased.

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