Global Veterinaria 8 (6): 665-669, 2012
ISSN 1992-6197

The Fatty Acids Profile in Mackerel (*Scomberomorus guttatus*) and its Shelf Life in Cold Storage at -18°C

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Abstract: This study was conducted to identify and determine the fatty acids profile in mackerel (*Scomberomorus guttatus*) meat as well as its changes during cold storage (-18°C). The only 15 of 30 types of authentic standard fatty acids were recognized in mackerel lipid. These saturated fatty acids (SFAs) were Myristic (C14:0), Palmitic (C16:0), Margaric (C17:0), Stearic (C18:0), Arachidic (C20:0), Tricosanic (C23:0) and Lignoseric (C24:0) acids. Also, unsaturated fatty acid (UFAs) as Palmitoleic (C16:1), Heptadecanoic (C17:1) and Oleic acids (C18:1) as well as poly unsaturated fatty acids (PUFAs) as Linoleic (C18:2 (n-6)), Linolenic (C18:3 (n-3)), Arachidonic (C20:4 (n-6)), Eicosapentanoic (C20:5 (n-3)) and Decosahexanoic (C22:6 (n-3)) acids. The SFAs, UFAs with one and more double bond were decreased 40.55, 60.07 and 66.56%, respectively ($P<0.05$) during cold storage (-18°C) for eight months. The obtained results of current study recommended that the shelf life of this species is five months in cold steerage (-18°C).

Key words: Fatty Acids · Shelf Life · Mackerel Meat

INTRODUCTION

Fish meat has very high nutrition value. The lipids of their meat contain a high level of PUFA [1, 2]. The importance of many fishes have discussed due to their contents of ω-3 and ω-6 fatty acids (FAs). The consumption of fishes containing high levels of FAs is favorable for human health, it could be used to prevent diseases (malnutrition and heart disease), recover more quickly from disease due to ω-3 and ω-6 UFAs [3]. Essential FAs (EFAs) cannot produce by human body, therefore these FAs have to be provided through human diets for keeping healthy human body. The benefits effects of fish consumption are depending on both fat content and the PUFA composition [4]. The PUFA composition of fishes may vary among species. But, this considers little attention [5].

The importance of PUFA on growth rate, nervous systems of human embryos and children as well as their degradation by auto oxidation during storage and processing [1] leads to continuous efforts to find out the best way to preserve and prolong shelf life of PUFA. The freezing and frozen storage have largely been used to retain fish dietary and nutritional properties [6]. But, the fish muscles undergo a number of changes which modify their structural and functional properties during frozen storage [7] and lead to unpleasant flavors [8].The rate and degree of changes are dependent upon the fish species, the presence or absence of activators and inhibitors and storage conditions [1].

The effect of frozen storage on the lipid composition of fishes has been investigated in some studies [4, 9]. In pre- and post-spawned hake, the polyenoics and ω-3 FAs changed during frozen storage [9, 10]. The damage of lipid fraction in whole jack mackerel (*Trachurus symmetricus murphyi*) showed that the lipid hydrolysis was the major damage during frozen storage [10].

The mackerel (*S. guttatus*) is commercially important fatty fishes that lives in epipelagic-neritic zone, in Persian Gulf and Sea of Oman, commonly processed as frozen fillets [11]. One of the characteristic of mackerel tissue is the amount of dark muscle which is soft and has desirable flavor in raw and cooked form. There are little study on quantity and quality of lipids and fatty acids identification of these fishes. Therefore this study was carried out to identify the quantity and quality of the fatty acids profile of Persian mackerel and their stability in cold storage at -18°C.

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MATERIALS AND METHODS

Twenty mackerel fishes (*S. guttatus*) were caught from Persian Gulf near to Khuzestan province (South-Iran) in this research. The length of the specimens was in the 55-705 cm range and the average weight of the specimens of mackerel was 2.70 kg. The samples were immediately transported under ice powder in foamed polystyrene self-draining boxes within 45 minutes to laboratory and then frozen by plate freezer at -30°C in order to minimize the effects of biochemical changes during transportation from sea to laboratory. They kept in freezer at -18°C after freezing.

Schedule of Sampling: Fish samples were used to determine the proximate composition, fatty acids and stability of fatty acids profile at -18°C. Analysis of frozen fish was undertaken as fresh (0) and after, 30, 60, 90, 120, 150, 180, 210 and 240 days under cold storage at -18°C. The proximate composition of samples was carried as fresh after thawing of the fishes.

Two fishes from both sexes were thawed in a refrigerator (at a temperature of 4±1°C) overnight and deboned, minced and the minced meat of each sex was blend with each other by laboratory mixer to obtain a homogeneous mixture. The samples of obtained mixture were used for determination the proximate composition and fatty acid identification.

Proximate Composition: The water and crude ash contents were determined in an oven at 103°C and 550°C, respectively, until the weight became constant. The crude protein was determined by Kjeldahl method [12]. The lipids were extracted by the method of Bligh and Dyer [13].

Fatty acid Analysis: According to given time table the samples were prepared and the total lipids were extracted by chloroform-methanol (1:1, v/v) and estimated gravimetrically [13]. The fatty acids in the total lipids were esterifies into methyl esters by saponification with 0.5 N-methanolic NaOH and transesteriried with 14% BF3 (Trifluoride Bore) (w/v) in methanol [12]. The fatty acid methyl esters were analyzed on a Hewlett Packard 6890 Gas Chromatograph (GC) equipped with a Flame Ionization Detector (FID). The esters were separated on a BPX-70 column (120 m ×0.25 mm i.d.). Column injector and detector temperatures were 285 and 320°C, respectively. The carrier gas was nitrogen (flow 0.6 ml min^{-1}). Identification of UFAs was performed by comparison to retention time of authentic standards. All the experiments were carried out in triplicate.

Statistical Analysis: All the data were subjected to analyses of variance (one-way ANOVA), at the $P<0.05$ confidence level using Duncan's multiple range test [14].

RESULTS AND DISCUSSION

Proximate Comparison: The results of proximate composition of mackerel meat are given in Table 1. The results showed that moisture level of mackerel meat is relatively high (72.75%). The crude lipids and protein levels are 7.50 and 15.99%, respectively and crude ash (3.76%) was in low levels of mackerel meat. Based on proximal analysis, water content in mackerel was near to other study which moisture in mackerel (*S. commersoni*) was 73.32% [4] and in Spanish mackerel 71.71% [15]. This showed that difference in moisture not varied in individual fish spices. The levels of lipid in fish tissue fluctuate based on species, ranging from lean fish (< 2% total lipid) such as cod, haddock and pollack, to high lipid species (8-20% total lipid) such as herring, mackerel and farmed salmon. In addition to species, sex,

Table 1: Approximate composition of mackerel (*S. guttatus*)

Approximate composition	%
Protein	15.99
Lipid	7.50
Moisture	72.75
Ash	3.76

Table 2: Identification of fatty acids of mackerel (*S. guttatus*) ($mg^{-1}g$)

Fatty acids	$mg^{-1}g$	Retention time (min)
C14:0 (Myristic acid)	0.33^{e}+0.04	4.01
C16:0 (Palmetic acid)	2.05^{a}+0.02	5.93
C16:1 (Palmitoleic acid)	0.51^{d}+0.03	6.51
C17:0 (Margaric acid)	0.29^{e}+0.01	7.07
C17:1(Heptadanoic acid)	0.07^{h}+0.02	7.42
C18:0 (Stearic acid)	0.98^{c}+0.03	8.12
C18:1 (Oleic acid)	0.97^{c}+0.76	8.55
C18:2 (n-6) (Linoleic acid)	0.18^{fg}+0.04	9.34
C18:3 (n-3) (Linolenic acid)	0.08^{gh}+0.03	10.47
C20:0 (Arachidic acid)	0.08^{gh}+0.00	11.18
C20:4 (n-6) (Arachidonic acid)	0.17^{fgh}+0.03	14.42
C20:5 (n-3) (Eicosapentaenoic acid)	0.28^{ef}+0.02	16.52
C23:0 (Tricosanoic acid)	0.12^{gh}+0.01	20.37
C24:0 (Lignoceric acid)	0.02^{h}+0.00	22.18
C22:6 (n-3) (Docosahexaenoic acid)	1.34^{b}+0.03	22.87
SEM	0.22	-
P value	<0.0001	-

Means with different superscripts in the same column are significantly different ($P<0.05$). SEM: Standard Error of Means

diet, seasonal fluctuation and tissue influence lipid levels [16, 17]. For example 20% in comparison with 4% lipid was found in the dark and white muscle of mackerel. A total lipid level of 7.5% has been suggested as a cut-off point between low and medium fat fish in this study. It is well known that oily fish are particularly susceptible to lipid oxidation, rancidity development and FAs reduction [16]. Therefore, it is seems that mackerel meat potential to FAs reduction is mediated between fish species. Also, it has been reported that low-fat fishes have higher water content [5], as observed in this study which was confirmed by other research [4].

The protein content of mackerel (15.99%) was lower than amount of protein (20.42%) in other study [15]. Moreover, the ash content (3.76%) was higher than in other result (1.24%) [15]. These variations may was related to sex and age of fishes.

Fatty Acids Composition: The identified mackerel meat fatty acids ($mg^{-1}g$) and retention times (min) are given in Table 2. The results showed that only 15 of 30 types of authentic standard FAs were identified in mackerel lipid in this investigation according to retention time. These were Myristic (C14:0), Palmitic (C16:0), Margaric (C17:0), Stearic (C18:0), Arachidic (C20:0), Tricosanic (C23:0) and Lignoseric (C24:0) acids which are saturated fatty acids. Also, eight UFAs were recognized, mono unsaturated fatty acid (MUFAs), Palmitoleic (C16:1), Heptadecanoic (C17:1) and Oleic acids (C18:1) as well as PUFAs, Linoleic (C18:2 (n-6)), Linolenic (C18:3 (n-3)), Arachidonic (C20:4 (n-6)), Eicosapentanoic (C20:5 (n-3)) and Decosahexanoic ((C22:6 (n-3)) acids. The higher levels of FAs were related to C16:0 (2.05 $mg^{-1}g$) ($P<0.05$). Also, the low level of C17:1, C18:3 (n-3), C20:0, C20:4 (n-6), C23:0 and C24:0 (0.07, 0.08, 0.08, 0.17, 0.12 and 0.02 $mg^{-1}g$) were found in FAs profile of mackerel meat ($P<0.05$). The FAs profile of fish varies quite considerably between and within species and many factors such as sex, diet, seasonal fluctuation and tissue lipid levels influence Fas profile [16]. Thus, observed FAs variations in this investigate can be due to mentioned factors.

The changes of SFAs and MUFAs during time table of 240 days of storage at -18°C are shown at Table 3. An obvious decrease trend was found in SFAs during the period of storage (fresh meat to 240 days of storage meat) ($P<0.05$).

The higher reduction was found by C24:0 which was virtually reached from 0.02 to 0.00 ($P<0.05$). The results of present study have shown that SFAs were very slowly decreasing during the first months of storage, but the rate of oxidation were increase after six months of storage and the total amount of SFAs from 3.86 $mg^{-1}g$ reached to 2.29 $mg^{-1}g$ after eight month of storing at -18°C. On the other hand, total FAs of mackerel fresh fish (100%) was reached to 59.45% that means 40.55% reduction occurred after eight months storage at -18°C. The Palmitic acid (C16:0) was the major FAs among the SFAs and during the whole period [4]. The same result was obtained for *Channa* spp [17] and mackerel [4].

The reduction of MUFAs was much faster as expected from SFAs. The 60.97% reduction during eight months of storage at -18°C was observed by MUFAs. The individual decreases of MUFA were 0.97 to 0.40, 0.07 to 0.02 and 0.51 to 0.18 $mg^{-1}g$ for C18:1, C16:1 and C17:1, respectively. Oleic acid (C18:1) was the major fatty acid among the MUFAs. The same result was shown in *Channa* spp. [17], gilthead sea bream (*Sparus aurata*), sea bass (*Dicentrarchus labrax*) [18] and mackerel [4].

Table 3: The occurred modifications of saturated and mono unsaturated fatty acids during frozen storage at -18°C ($mg^{-1}g$)

Fatty acids	30 days	60 days	90 days	120 days	150 days	180 days	210 days	240 days	P value
C14:0	0.29^{a}+0.00	0.29^{a}+0.00	0.23^{b}+0.00	0.23^{b}+.01	0.23^{b}+0.03	0.22^{b}+0.07	0.21^{b}+0.05	0.20^{b}+0.02	<0.0001
C16:0	2.01^{a}+0.01	1.70^{b}+0.02	1.65^{c}+0.01	1.64^{c}+0.03	1.54^{d}+0.02	1.51^{d}+0.02	1.33^{e}+0.01	1.31^{e}+0.02	<0.0001
C17:0	0.28^{a}+0.07	0.23^{b}+0.05	0.22^{c}+0.02	0.22^{c}+0.02	0.21^{c}+0.02	0.21^{c}+0.03	0.21^{c}+0.02	0.18^{d}+0.07	<0.0001
C18:0	0.83^{a}+0.07	0.74^{b}+0.07	0.71^{c}+0.06	0.63^{d}+0.10	0.56^{e}+0.09	0.53^{f}+0.09	0.49^{g}+0.01	0.48^{g}+0.09	<0.0001
C20:0	0.08^{a}+0.02	0.07^{b}+0.01	0.05^{c}+0.04	0.03^{d}+0.01	0.03^{d}+0.00	0.03^{d}+0.01	0.03^{d}+0.02	0.02^{e}+0.00	<0.0001
C23:0	0.12^{a}+0.07	0.11^{a}+0.00	0.11^{a}+0.01	0.07^{b}+0.01	0.07^{b}+0.01	0.07^{b}+0.06	0.05^{b}+0.01	0.05^{b} +0.02	0.0111
C24:0	0.02^{a}+0.01	0.01^{b}+0.00	0.00^{c}+0.00	0.00^{c}+0.00	0.00^{c}+0.00	0.00^{c}+0.00	0.00^{c}+0.00	0.00^{c}+0.00	<0.0001
Total	3.60	3.15	2.99	2.82	2.65	2.58	2.32	2.29	
C16:1	0.50^{a}+0.07	0.47^{b}+0.03	0.42^{c}+0.02	0.42^{c}+0.03	0.40^{d}+0.07	0.31^{e}+0.04	0.22^{f}+0.00	0.18^{g}+0.01	<0.0001
C17:1	0.06^{a}+0.07	0.06^{a}+0.05	0.06^{a}+0.06	0.06^{a}+0.01	0.05^{b}+0.17	0.05^{b}+0.21	0.04^{b}+0.37	0.02^{c}+0. 47	<0.0001
C18:1	0.91^{a}+0.37	0.88^{b}+0.10	0.85^{c}+0.08	0.75^{d}+0.07	0.74^{e}+0.11	0.51^{f}+0.12	0.41^{g}+0.17	0.40^{h}+0.06	<0.0001
Total	1.46	1.41	1.32	1.22	1.19	0.87	0.67	0.60	

Means with different superscripts in the same raw are significantly different ($P<0.05$).

Table 4: The changes of polyunsaturated fatty acids during 240 days of storage at -18°C ($mg^{-1}g$)

Fatty acids	30 days	60 days	90 days	120 days	150 days	180 days	210 days	240 days	P value
C18:2 (n-6)	0.17^{a}+0.07	0.17^{ab}+0.03	0.16^{b}+0.02	0.16^{b}+0.02	0.16^{b}+0.03	0.09^{c}+0.02	0.09^{c}+0.02	0.06^{d}+0.07	<0.0001
C18:3 (n-3)	0.06^{a}+0.60	0.05^{b}+0.50	0.04^{c}+0.02	0.04^{c}+0.01	0.04^{c}+0.00	0.04^{c}+0.01	0.03^{d}+0.02	0.02^{e}+0.02	<0.0001
C20:4 (n-6)	0.17^{a}+0.06	0.16^{b}+0.02	0.16^{b}+0.01	0.14^{c}+0.01	0.13^{c}+0.05	0.13^{c}+0.04	0.10^{d}+0.00	0.10^{d}+0.00	<0.0001
C20:5 (n-3)	0.27^{a}+0.01	0.23^{b}+0.00	0.23^{b}+0.00	0.18^{c}+0.07	0.17^{c}+0.07	0.15^{c}+0.00	0.11^{d}+0.07	0.10^{d}+0.50	0.0121
C22:6 (n-3)	1.31^{a}+0.05	1.30^{a}+0.017	1.21^{b}+0.06	0.80^{c}+0.07	0.78^{cd}+0.77	0.71^{d}+0.42	0.59^{e}+0.14	0.41^{e}+0.07	0.0277
Total (%)	96.79	92.76	87.52	64.17	62.23	54.51	44.27	33.44	

Means with different superscripts in the same raw are significantly different (P<0.05).

The changes of PUFAs contents ($mg^{-1}g$) are shown at Table 4 during meat storage for 240 days at -18°C. The results of variation showed that the higher reduction of ω-3 content was related to C22:6 (n-3) and the lower one was belonged to C18:3 (n-3). Moreover, the C18:2 (n-6) had maximum rate and lower variation between ω-6 FAs (P<0.05). As another word, the changes of ω-3 and ω-6 were 55.67% decreases after eight months. The individual PUFAs reduction showed that C22:6 (n-3) reduced from 1.34 to 0.41 $mg^{-1}g$ and C20:5 (n-3) from 0.28 to 0.10 $mg^{-1}g$ (P<0.05). The most reductions were related to C18:3 (n-3) (0.08 to 0.02 $mg^{-1}g$) and C20:4 (n-6) (0.17 to o.10 $mg^{-1}g$) after frozen storage (at -18°C) period. The results showed that UFAs were more than SFA (SFA<PUFA+MUFA) while, during frozen storage, UFAs decreased in contrast with SFA. Distribution of FAs in mackerel was as SFA> MUFA>PUFA. This is consistent with channel catfish [19], seal [20] and mackerel [4]. The species of fish is affected PUFA composition [17] but, the amount of PUFA is affected by diet [21]. The C20:5 (n-3) and C22:6 (n-3) were major in total PUFAs in this study. The same results were observed in gilthead sea bream (*Sparus aurata*) and sea bass (*Dicentrarchus labrax*) [18]. The lowest amount of ω-3 compounds were found in C18:3 (n-3). The same results were found for seal [20], *Channa* spp. [17] and mackerel [4].

The differences in the FAs composition of the fish species lipids had a critical function in the hydroperoxides formation. The oxidative changes in frozen fish lipids may be caused the occurrence of radicals [22]. These radicals are easily formed in mackerel, because of more PUFA. It is illustrated that mackerel is very susceptible to lipid peroxidation due to its high PUFA content during frozen storage [22].

The ω-3: ω-6 ratio suggested as a useful indicator for nutritional values of fish oils. A ratio of 1:1 to 1:5 would constitute a healthy human diet [5, 17].This study has shown that marine fish are richer in ω-3 than ω-6 PUFAs. The decrease in mackerel showed that the nutritional value of these fish has been lost during frozen storage. The PUFA/ SFA ratio reveals that marine fish are a good source of PUFA related to SFAs. This ratio obtained was less than 1 in mackerel. Any decrease of PUFA relative to SFA leads to a significant decrease of this ratio (58.50%) during frozen storage. The negative relationship between this ratio and storage time showed that oxidation mechanisms are active during frozen storage. These functions lead to reduction in nutritional value of fish meat after storage period. On the other hand, the obtained results have demonstrated that in Persian Gulf mackerel, the least changes belong to SFA then followed PUFAs and finally MUFAs during the frozen storage at -18°C.

CONCLUSION

The lipid content as a result of frozen storage conditions (time and temperature) were decreased which reflected on FAs content. Determination of fatty acids composition indicated that mackerel meat has equal amounts of saturated and unsaturated fatty acids and have high rate of ω-3 and ω-6 fatty acid groups. Palmitic acid (C16:0) is more abundant fatty acid. Lipid deterioration is the main cause of the low shelf life of FAs due to oxidation and hydrolysis. The quality of mackerel meat was decrease during preservation period. This species of fish if immediately frozen can be usable up to 5 months. According to nutritional value of this Persian Gulf species, it is not classified as an excellent fishes.

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