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Fatty Acid and Proximate Composition of Farmed Great Sturgeon (*Huso huso*) Affected by Thawing Methods, Frying Oils and Chill Storage

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Abstract

The objective of this study was to investigate the changes in proximate and fatty acid composition of farmed great sturgeon (*Huso huso*) as influenced by thawing methods, frying oils and chill-storage. Slices were frozen for one month (-18ºC), thawed by refrigerator (RT, 4ºC) or microwave (MT), fried in canola, soybean and hydogenated vegetable oils and then analysed immediately after frying or after 3 days chilling (4ºC). Frying by canola oil decreased the content of polyunsaturated fatty acids (PUFAs) while frying in soybean and hydrogenated vegetable oil had the opposite effects. Frying decreased the contents of EPA, DHA and n-3 fatty acids. MT slices had lower n-3 essential polyunsaturated fatty acid contents than RT slices after

frying. The ratio of n-6/n-3 fatty acids increased after frying and exceeded the recommended ratio in the slices fried by hydrogenated vegetable and soybean oils. The results indicated that RT slices had the fatty acid profile closer to the control samples compare to MT slices. Moreover proximate and fatty acid composition of slices were affected by frying oils while chill storage had little effect.

Keywords: Thawing, Frying, Chill storage, Fatty acid composition, Farmed great sturgeon

Introduction

Freezing is a widely used and effective method for preserving foods. During frozen storage some quality deterioration occurs in food particularly texture, flavor and color. The extent of quality loss depends on many factors such as freezing rate, thawing methods, temperature fluctuations [1]. Frozen foods are commonly thawed before processing or cooking which can lead to chemical, physical and microbiological damages in food. As thawing generally takes longer than freezing, the purpose of thawing process is to preserve the original quality of the food [2]. Rapid thawing at low temperatures can prevent quality loss of food during production. Boonsumrej et al. [3] reported that refrigerator thawing and microwave thawing are two satisfactory methods for thawing foods. Xia et al. [2] noted that pork muscle thawed by refrigerator had the least quality loss and its physicochemical properties were more close to fresh muscle than the other thawing methods.

Fish meat is seldom eaten as raw, but is prpared in diffent ways before consumption. Frying is generally the most common method of food preparation. It gives unique organoleptic characteristics to foods such as flavor, texture and appearance which can improve the palatability of fried foods [4]. During frying of food, water is replaced by the frying oil and as a consequence, oil becomes an important component of the fried food [5]. Different frying oils are available in the markets and used to prepare foods. The effects of different cooking oils on the essential omega-3 fatty acid of fish meat have been investigated [6, 7].

Omega-3 polyunsaturated fatty acids are highly important for human health as these fatty acids have beneficial role on reducing artherosclerosis, prevention and treatment of numerous disorders like cardiovascular disease and others [8]. Fish and shell-fish are the main source of omega-3 fatty acids and humans obtain main parts of these fatty acids through consuming aquatic products [9]. Omega-3 polyunsaturated fatty acids are very susceptible to oxidation which not only affect the sensory attributes of the foods, but also contributes to many disease in humans [10].

Sturgeons perhaps are one of the most important wildlife commodities on earth. They occupied diverse habitats of the northern hemisphere and produce the most famous luxury food product, caviar [11, 12]. Sturgeon meat is rich in protein of good

biological value. It has medium fat content and is also rich in essential ions and vitamins [13]. Great sturgeon (*Huso huso*) is a good candidate for aquaculture due to its fast growth, big size and tasty flesh [14]. Although the effects of thawing methods on physicochemical characteristics of muscle food has been extensively studied, little information has been reported (particularly for farmed sturgeon species) on the changes of essential omega-3 fatty acids and omega-6/omega-3 ratio as a result of combined effects of thawing methods, frying by different oils and chill storage. Therefore the objective of the present study was to elucidate the changes in proximate and fatty acid composition of farmed great sturgeon (*Huso huso*) as influenced by thawing methods, frying oils and chill storage.

Materials and methods

Sample preparation

Farmed male great sturgeons (average weight= 5 kg, n= 3) were purchased from a local sturgeon farm in northern Iran (SaeeGostar, Sari, Mazandaran). The fish were reared in concrete circular tanks (8 m diameter, 1.5 m depth) at 30 kg/m^3 in density and fed 2% of body weight with trout commercial diets contained 40% protein and 15% lipid. The major fatty acids of the diet were C16:0 (18.83%), C18:0 (5.04%), C16:1 (3.17%), C18:1 (27.78%), C18:2 (27.27%), C18:3 (8.99%), C20:5 (0.59%) and C22:6 (3.47%). At the farm, the fish were beheaded, eviscerated, washed, placed in ice (fish to ice ratio of 1:2 (w/w)) and transported to laboratory. At laboratory the fish were deskinned and washed with cold tap water to remove adhering blood and slime. The fish were then cut into slices of approximately 1 cm thickness. All slices were vacuum-packed and stored at -18ºC for one month. The frozen slices were thawed using refrigerator (RT, 4ºC) or microwave (MT, Major, MCO 3515, Iran). RT and MT slices were didived into four homogenour groups.

Frying and chill storage

For performing the experiment, each above mentioned groups was divided into two groups: group 1: samples were analyzed after frying (day 0); group 2: samples were chilled-stored at 4ºC for 3 days and then analyzed (day 3). All slices (except than non-fried fresh group) were fried for 4 min at 160 ºC in a deep-fryer (Hamilton, HDF-510, Iran). The oil/slice ratio was 2:1. After frying slices were allowed to be air cooled for 2 min prior to analysis. The oils used for frying were canola (SFA: 5.66%, MUFA: 65.17%, PUFA: 25.28%, n-3: 7.76%, n-6: 17.52%, n-6/n-3: 2.26), soybean (SFA: 13.39%, MUFA: 21.44%, PUFA: 63.38%, n-3: 7.36%, n-6: 56.02%, n-6/n-3: 7.61), and hydrogenated vegetable oil (SFA: 23.77%, MUFA: 29.37%, PUFA: 45.11%, n-3: 4.35%, n-6: 40.76%, n-6/n-3: 9.37) purchased from a local market.

Proximate composition

Moisture was determined by drying the samples in an oven (Heraeus, D-63450, Hanau, Germany) at 105°C to a constant weight [15]; lipid was extracted according to Bligh and Dyer [16]. Ash was determined by incineration in a muffle furnace (Isuzu, Tokyo, Japan) at 600 °C for 3 h [15]; crude protein was determined by the Kjeldahl method (N× 6.25) using an automatic Kjeldahl system (230-Hjeltec Analyzer, Foss Tecator, Höganäs, Sweden) [15].

Lipid extraction

Lipid was extracted according to the method of Bligh and Dyer [16]. Fifty g of sample were homogenized in a blender for 2 min with a mixture of 50 ml chloroform and 100 ml methanol. Then 50 ml of chloroform were added and further homogenized for 30 sec. Finally 50 ml of distilled water were added to the mixture and blended for 30 sec. The homogenate was centrifuged (Avanti J-E, BECKMAN COULTER, Inc., USA) at 3000 rpm for 15 min at 4°C. Supernatant was then transferred into a separating flask and the lower phase (chloroform phase) was drained off into a 250 ml Erlenmeyer flask containing 4 g anhydrous sodium sulfate and shaken vigorously. The solution was then filtered through a Whatman No. 4 filter paper into a round-bottom flask. Rotary evaporator (Rotavapor R-114, BÜCHI, Flawil, Switzerland) was used for solvent evaporation at 25°C.

Fatty acid analysis

Fatty acid methyl ester was prepared as follows: Lipid samples (1 g) were diluted with 2 ml of 2 M potassium hydroxide in methanol followed by the addition of 7 ml n-hexane in a sealed tube. The mixture was then shaken using a vortex for 1 min and left for about 20 min in a water bath (temperature 50-55ºC) until it was separated into two phases. From top layer, fatty acid methyl ester was then taken for analysis by using Trace GC (Thermo Finnigan, Italy). The GC conditions were as follows: capillary column (Bpx-70, 60 m, 0.32 mm, i.d. 0.25 μm); the split ratio of 90:1; injection port temperature of 250 °C; flame ionization detector temperature of 270 °C. Oven temperature was set at 195 ºC for 75 minutes. Flow rate of carrier gas (helium) was 1 mL min^{-1} and the makeup gas was N_2 (30 ml/min). The sample size injected for each analysis was 1 μL. The data are expressed as g/100 g of total fatty acids.

Statistical analysis

Data were analyzed by one-way analysis of variance (ANOVA) and Duncan's multiple range test ($P < 0.05$) by SPSS 16.

Results and discussion

Proximate composition

Proximate composition of farmed great sturgeon raw slices is shown in Table 1. Moisture accounts for 70% of the slices while protein; lipid and ash contents were 18-19%, 3-4.5% and 1.9-2.8% respectively. The content of protein in farmed great sturgeon is similar to protein content of other farmed sturgeon species in the ranges of 15.2-21% [13, 17, 18, 19, 20, 21]. Sturgeon is medium-fat fish species with lipid content between 5-15% [13]. The content of lipid in farmed great sturgeon (3-4.5%) is similar to lipid content of other sturgeon species such as farmed white sturgeon (*Acipenser transmontanus*) with 3.4% lipid [21] and Russian sturgeon (*A. guldenstaedtii*) with 4-7% lipid [18] but is lower than that of other species such as farmed *Acipenser spp.* with 7.63% lipid [13] and Siberian sturgeon (*A. baerii*) 8-11.6% [20] and sturgeon hybrid (*A. baerii* × *A. medirostris*) with 6.4% lipid [19]. After frying, the contents of moisture decreased while lipid increased ($P<0.05$) and similar results was reported by Garcia-Arias et al. [22] for sardine (*Sardina pilchardus*), Gokoglu et al. [23] for rainbow trout (*Oncorhynchus mykiss*), Weber et al. [8] for silver catfish (*Rhamdia quelen*) and Ersoy and Özeren [24] for African catfish (*Clarias gariepinus*). Lipid increase after frying is due to oil penetration on the food after water is partially lost by evaporation [25]. However there was no difference in moisture and lipid content of the slices thawed by either refrigerator (RT) or microwave (MT) after frying. The content of protein and ash did not change after frying by different oils and no significant difference was observed for protein and ash content between RT and MT slices (Table 1).

Fatty acid composition

Fatty acid composition of farmed great sturgeon flesh is presented in Tables 2 and 3. Linoleic acid (C18:1), linolenic acid (C18:2) and palmitic acid (C16:0) were the main fatty acids of raw flesh (Table 2). Monounsaturated fatty acids were the most dominant class of fatty acids followed by polyunsaturated and saturated fatty acids and similar pattern was reported for sturgeon hybrids *A. schrenckii*× *Huso dauricus* and *A. baerii*× *A. medirostris* [19, 26]. N-6 fatty acids were higher than n-3 fatty acids gave an N-6/n-3 ratio of 2.4-2.8. The contents of eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) contents of farmed great sturgeon flesh in this study (1.45 and 5 g/100 g of total fatty acids respectively) were lower than those of other farmed sturgeon species such as Siberian sturgeon (*Acipenser baerii)*, Adriatic sturgeon (*A. naccarii)* and white sturgeon *(A. transmontanus)* with 4.8-6.54 g/100 g EPA and 8.7-9.7 g/100 g DHA [27]. Lower contents of EPA and DHA in the farmed great sturgeon in this study coincided with the lower contents of these fatty acids in the diet (0.59 and 3. 47 g/100g of total fatty acids respectively).

The effects of frying of farmed great sturgeon slices by different oils on the fatty acid composition are presented in Tables 2 and 3. The content of saturated fatty acids decreased after frying in canola and soybean oil, however it remained unchanged when slices fried by hydrogenated vegetable oil. Fried slices by canola oil, had the increase in monounsaturated fatty acids. This increase is due to the higher amounts of

oleic acid (65.17 g/100 g of total fatty acids) in canola oil. However after frying by soybean and hydrogenated vegetable oils, the content of MUFAs decreased. Fatty acid composition of soybean and hydrogenated vegetable oils was dominated by high amounts of polyunsaturated fatty acids mainly due to the presense of high content of linoleic acid in these oils and the content of this fatty acid exhibited significant increase after frying and instead when canola oil was used for frying, the content of

Table 1. Proximate composition of raw and fried slices of farmed great sturgeon thawed by refrigerator (RT) and microwave (MT).

		Raw slices		Canola		soybean		Hydrogenated vegetable oil	
		Day 0	Day 3	Day 0	Day 3	Day 0	Day 3	Day 0	Day 3
L**	RT	4.25±0.91b	3.75±1.34b	13.60±1.69a	12.75±0.49a	12.45±3.7a	11.10±3.81a	12.00±1.69a	11.05±2.61a
	MT	4.50±0.42b	3.30±0.98b	11.95±0.49a	12.10±0.42a	12.35±0.49a	11.95±2.19a	11.25±3.46a	12.40±0.42a
P	RT	18.45±0.49	18.15±0.49	18.10±1.31	18.45±0.77	17.55±0.21	18.30±0.70	18.40±0.56	18.60±0.56
	MT	18.90±0.28	18.35±0.49	18.15±0.21	17.85±0.07	18.70±0.56	18.30±0.70	18.50±0.56	18.55±0.63
M	RT	70.75±1.34a	71.90±0.84a	62.55±1.48b	62.05±0.77b	62.20±0.28b	62.15±0.07b	61.85±0.49b	61.15±0.35b
	MT	70.45±2.04a	70.19±1.14a	62.75±1.75b	63.05±0.07b	62.20±0.28b	61.95±0.35b	62.30±1.13b	62.45±1.32b
A	RT	1.90±0.28	2.15±0.07	1.89±0.14	2.65±0.63	1.95±0.07	2.45±0.07	2.00±0.28	2.20±0.42
	MT	2.30±0.14	2.75±0.35	2.25±0.08	2.00±0.42	3.02±1.30	2.410±0.28	2.50±0.56	2.55±0.06

* Means with the same superscripts within the same row were not significantly different ($P > 0.05$). ** L= Lipid, P= Protein, M= Moisture, A= Ash.

Table 2. Fatty acid compoition (g/ 100 g of total fatty acids) of raw and fried slices of farmed great sturgeon thawed by refrigerator (RT) and microwave (MT).

		Raw slices		Canola		soybean		Hydrogenated vegetable oil	
		Day 0	Day 3	Day 0	Day 3	Day 0	Day 3	Day 0	Day 3
C14:0	RT	1.05	1.00	0.49	0.69	0.56	0.87	0.40	0.78
	MT	1.14	0.79	0.70	0.55	0.55	0.63	0.44	0.64
C16:0	RT	16.44	17.76	9.55	12.00	14.78	14.22	16.55	17.68
	MT	16.32	18.49	11.85	10.21	13.60	13.83	18.11	18.27
C16:1	RT	2.98	2.78	1.33	1.86	1.56	1.55	0.75	1.71
	MT	3.02	2.17	1.84	1.47	1.39	1.63	0.68	1.31
C18:0	RT	3.18	3.33	2.80	2.68	3.99	3.29	4.84	4.24
	MT	2.91	4.09	2.73	2.71	3.77	3.64	5.47	4.63
C18:1	RT	34.14	34.44	50.43	45.58	30.14	32.17	29.66	33.20
	MT	35.95	32.22	46.29	49.33	29.17	29.54	30.96	32.56
C18:2	RT	24.00	21.63	19.74	21.45	35.16	35.11	33.89	29.69
	MT	22.47	24.25	20.97	21.03	38.50	36.35	35.21	31.95
C18:3	RT	2.46	2.20	2.76	3.18	4.04	4.18	3.12	3.11
	MT	2.55	2.35	2.64	2.74	4.67	4.34	3.42	3.27
C20:1	RT	0.75	0.96	0.40	0.65	0.77	0.50	0.35	0.49
	MT	0.57	1.19	0.41	0.36	0.40	0.40	0.35	0.28
C20:5	RT	1.50	1.44	0.85	0.97	0.94	0.78	0.59	0.89
	MT	1.42	1.45	0.88	0.68	0.73	0.79	0.45	0.63
C22:6	RT	5.08	5.30	2.56	3.23	3.53	2.77	2.12	2.83
	MT	4.29	4.88	2.90	2.41	2.46	2.51	1.66	2.20

Table 3. Major class of fatty acids (g/ 100 g of total fatty acids) of raw and fried slices of farmed great sturgeon thawed by refrigerator (RT) and microwave (MT).

		Raw slices		Canola		soybean		Hydrogenated vegetable oil	
		Day 0	Day 3	Day 0	Day 3	Day 0	Day 3	Day 0	Day 3
Σ SFA	RT	20.67	22.00	12.84	15.37	19.33	18.38	21.79	22.7
	MT	20.37	23.37	15.28	13.47	17.92	18.10	24.02	23.54
Σ MUFA	RT	37.87	38.18	52.61	48.09	32.47	34.22	30.76	35.40
	MT	39.54	35.58	48.54	51.16	30.96	31.57	31.99	34.15
Σ PUFA	RT	33.04	30.57	25.91	28.83	43.67	42.84	39.72	36.52
	MT	30.73	32.93	27.39	26.86	46.36	43.99	40.74	38.05
Σ N-3	RT	9.04	8.94	6.17	7.38	8.51	7.73	5.83	6.83
	MT	8.26	8.68	6.42	5.83	7.86	7.64	5.53	6.36
Σ N-6	RT	24.00	21.68	19.74	21.45	35.16	35.11	33.89	29.69
	MT	22.47	24.25	20.97	21.03	38.50	36.35	35.21	31.95
N-6/n-3	RT	2.65	2.42	3.20	2.90	4.13	4.54	5.81	4.34
	MT	2.7	2.80	3.26	3.60	4.90	4.80	6.36	5.02

this fatty acid decreased. Frying decreased the contents of EPA, DHA and n-3 fatty acids in all samples indicating that most of the fatty acids of the fried slices of farmed great sturgeon become similar to those of frying oils and similar results were reported for Indo-Pacific king mackerel (*Scomberomorous guttatus*) by Bakar et al. [28] and sardine (*Sardina pilchardus*) by Garcia-Arias et al. [22]. During frying process, an exchange of oil between the food and the culinary oils takes place, thereby altering the lipid composition of fried foods to resemble to the oils used [5, 6, 29]. The contents of EPA, DHA and total n-3 fatty acids were higher in RT slices fried by different oils when compared to MT slices. Boonsumrej et al. [3] and Xia et al. [2] have also noted that shrimp and pork muscle slices thawed by refrigerator had the quality closer to fresh samples when compared to microwave-thawed samples. Chill storage of fried fish is also a common practice which may have an influence on its nutritional quality [28]. Proximate composition of raw slices, fatty acid composition remained stable and did not change significantly after 3 days of chill storage. The result is in agreement with those of Pirini et al. [30] for sea bass (*Dicentrarchus labrax*) and Senso et al. [8] for farmed gilthead sea bream (*Sparus aurata*) who reported no changes in fatty acid composition following chill storage of fillets.

N-6/n-3 ratio is an important determinant of health and lower ratio is desirable in reducing the risks of chronic diseases. Fish species are the rich source of n-3 essential fatty acids such as EPA and DHA which are very potent in lowering the many risk factors associated with chronic diseases and disorders in human body [9]. N-6/n-3 fatty acid ratio of raw farmed great sturgeon slices was in the ranges of 2.4-2.8. After frying in hydrogenated vegetable and soybean oils, this ratio increased significantly and exceeded the recommended ratio of 2:1-4:1 for human health as suggested by

Pepping [31] (Table 3). In Indo-Pacific king mackerel frying resulted in an increased N-6/n-3 ratio (from 0.54 in raw flesh to 1.2 in fried samples) [28] and similar result was noted by Garcia-Arias et al. [22] on sardine fillets.

In conclusion the results of this study indicated that slices thawed by refrigerator had the fatty acid composition closer to control samples compare to the slices thawed by microwave. Proximate and fatty acid composition of slices were affected by frying oils indicating fatty acid equilibrium between the culinary oil and slices. Moreover chill storage of fried slices had little effect on chemical composition of farmed great sturgeon.

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