

## Comparison of three different fish species from the Marmara Sea to fulfill daily EPA and DHA needs

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Received: October 2016

Accepted: March 2017

### Abstract

Fatty acid profile and proximate composition of horse mackerel (*Trachurus mediterraneus*), sole (*Solea solea*) and black scorpion fish (*Scorpaena porcus*) caught from the Marmara Sea were determined both in the summer (June-July-August) and winter (December-January-February) seasons. Fish is the best source of omega-3 ( $\omega$ -3) and contains plenty of omega-6 ( $\omega$ -6) fatty acids.  $\omega$ -3 fatty acids provide so many health benefits and help to protect against diseases. In this study, the lipid content of horse mackerel was higher than the other investigated fish species. Total  $\omega$ -3 and eicosapentaenoic acid (EPA) + docosahexaenoic acid (DHA) of horse mackerel were the highest in summer (1705 mg 100g<sup>-1</sup>) despite having higher lipid content in winter, which could be explained by the nourishment that fish takes in different seasons. Considering the recommended intake, EPA+DHA of horse mackerel was more than enough to meet the daily requirement. The  $\omega$ -3 content of black scorpion fish and sole were relatively low in comparison with horse mackerel in both seasons. The ratio of  $\omega$ -6: $\omega$ -3 fatty acids of all species was also very low (max. 0.358) which provides advantage in daily nutrition. Docosapentaenoic acid (DPA) and  $\alpha$ -linolenic acid (ALA) were also found higher in horse mackerel in winter. Instead of taking supplements, fish itself should be consumed to benefit all long chain  $\omega$ -3 polyunsaturated fatty acids (PUFA) and other nutrients. For a healthy diet, consuming horse mackerel routinely seems to fulfil EPA and DHA needs, besides having high quality protein.

**Keywords:** Fatty acids, Fish, Omega-3, EPA, DHA, Proximate composition

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## Introduction

The nutritional requirements of human beings are programmed according to the Paleolithic diet before the advent of agriculture. Forty thousand years ago our ancestors fed on fish, lean meat, fruit, vegetables, eggs and nuts. The Paleo diet differs from today's western dietary pattern in terms of lower saturated fat intake and balanced consumption of  $\omega$ -6 and  $\omega$ -3 fatty acids (1:1). On the other hand, in a western style diet the  $\omega$ -6 and  $\omega$ -3 fatty acid ratio is 15:1–16.7:1 which promotes many diseases (Simopoulos, 2002). According to the diet we have evolved on, fish seems the best option for eating healthy with a balanced ratio of fatty acids. Fish is a rich source of  $\omega$ -3 and  $\omega$ -6 polyunsaturated fatty acids (PUFA) and are also characterized by containing high-quality protein, vitamins and minerals (Prato and Biandolino, 2015). The major reference fatty acids of fish are  $\omega$ -3 fatty acids represented by eicosapentaenoic acid (EPA, C20:5  $\omega$ -3), and docosahexaenoic acid (DHA, C22:6  $\omega$ -3) (Tokuyama and Nakamoto, 2014). EPA and DHA have many benefits on various mechanisms in the body such as brain and visual system development (Serfaty and De Velasco, 2014) and induction and regulation of neuropathic pain (Tokuyama and Nakamoto, 2014). They are essential for serotonin production and have anti-depressant effects by providing neuroprotection and enhancing neurotransmission in the brain (Chandola and Tanna, 2014). Also, EPA and DHA have the potential in the remediation of memory deficits and are

beneficial to hippocampal learning and memory (Francis and Stevenson, 2014). Besides these benefits, EPA and DHA have protective effects for sudden cardiac deaths caused by arrhythmias and cardiovascular disease (Russo, 2009). Therefore, based on cardiovascular risk considerations, eating fish at least two times a week is recommended by the American Heart Association (Kris-Etherton *et al.*, 2002) and 250 – 500 mg intake of EPA and DHA daily is advised by EFSA (2012). Consuming fish regularly has great importance for fulfilling EPA and DHA needs. However, inadequate consumption of fish leads consumers to take fish oil supplements daily.

The Marmara Sea comprises one of the most important fishing areas of Turkish fisheries. In Turkey, total fisheries capture in 2015 was 345765 tons and catch from the Sea of Marmara relatively comprised a good part of the total catch (TUIK, 2016). Horse mackerel (*Trachurus mediterraneus*), sole (*Solea solea*) and black scorpion fish (*Scorpaena porcus*) are marketable fish species caught from the Marmara Sea and consumed throughout the year. According to data from the Turkish Statistical Institute for 2015 (TUIK, 2016) the total catches of horse mackerel, sole, and black scorpion fish were 14290.4, 328.0 and 143.2 tons, respectively.

The chemical and fatty acid profiles of a fish within the same species is variable and considerably depend on age, size, sex, habitat and fishing season (Sen, 2005). Feeding habits also determine the flesh composition and

fatty acid content (Periago *et al.*, 2005). Sole is a demersal and solitary fish, burrowing into muddy and sandy bottoms (2–200 m) and feeding on worms, mollusks and small crustaceans (Can and Bilecenoglu, 2005). Red scorpion fish is also demersal and solitary, but common among rocks and algae (20–500 m) and feeds on fish, crustaceans and mollusks. However, horse mackerel is an active pelagic and schooling fish, which migrates seasonally offshore and inshore (0–500 m), and feeds on fish, crustaceans, and cephalopods (Froese and Pauly, 2016). It is important to meet the EPA and DHA requirements by consuming fish itself, not only as supplements. The aim of this study was to determine the effects of seasons on the fatty acid profiles of three fish species living in different environments. The results provided information for suggesting the best season for the abundance of fatty acids of the investigated species in order to reveal their importance in human nutrition.

## Materials and methods

### *Fish sampling and preparation*

Horse mackerel (*T. mediterraneus*), sole (*S. solea*) and black scorpion fish (*S. porcus*) were caught from the Sea of Marmara. Sampling seasons were determined as summer (June–July–August) and winter (December–January–February). This study was conducted between 2013 and 2014 in two year periods. Total mid-caudal lengths of horse mackerel, sole and black scorpion fish were  $13.27 \pm 0.82$  cm;  $26.63 \pm 0.92$  cm and  $18.41 \pm 1.89$  cm

in winter and  $12.00 \pm 1.42$  cm;  $22.43 \pm 1.26$  cm and  $17.89 \pm 3.31$  cm in summer, respectively. Total weights were also measured which were  $26.38 \pm 4.81$  g;  $100.57 \pm 10.98$  g and  $121.54 \pm 39.00$  g in winter and  $16.90 \pm 5.86$  g;  $88.00 \pm 16.25$  g and  $120.25 \pm 73.16$  g in summer for horse mackerel, sole and black scorpion fish, respectively. For analyses sampling was carried out by using 7 kg fish and analyses for each fish were done in triplicate.

After transportation to the laboratory, fish were gutted and filleted. Fillets were homogenized separately in a homogenizer (Retsch, Grindomix GM200, Germany). For fatty acid analyses samples were immediately frozen and stored at  $-20^{\circ}\text{C}$  until use. Proximate composition analyses of the fish were done immediately.

### *Proximate composition*

Protein, lipid, moisture and ash content analyses were carried out in order to determine the proximate compositions of each sample. Protein and lipid content analyses were carried out according to the Association of Official Analytical Chemists (AOAC) method 955.04 (AOAC, 1998a) and AOAC method 991.36 (AOAC, 1998b), respectively. For the determination of moisture content, samples were dried to a constant weight at  $105^{\circ}\text{C}$  (Matissek *et al.*, 1992). Ash contents were determined with the method of AOAC 938.08 by burning samples completely to ash in a hot oven at  $550^{\circ}\text{C}$  (AOAC, 1998c). All results were expressed as  $\text{g } 100\text{g}^{-1}$ .

### Fatty acid analyses

Total lipids were extracted according to AOAC 991.36 (AOAC, 1998b) and fatty acid composition was determined by using gas chromatography (GC) (Perkin-Elmer Claurus 500 GC, Autosystem XL, FID, GC Software Turbochrom 4.1, Awenuid-Shelton, DE, USA) to analyze their methyl esters according to the method modified from Ichihara *et al.* (2002). GC conditions were as follows: Column SGE BPX70 (SGE Analytic Science, Australia) 60m x 0.25 mm ID x 0.25  $\mu$ m, injection volume 0.5  $\mu$ l, injection temperature 220°C, air 450 mL/min, H<sub>2</sub> 45 mL/min, flame ionization detector (FID) temperature 240 °C and column temperature, 120 °C for 5 min, programmed at 5 °C min<sup>-1</sup> up to 240 °C for 15 min (total program process time was 45 min). Fatty acids were identified by comparison of their retention times with a known reference material (Menhaden Fish Oil, 47116 Supelco) and authentic standards (Supelco 37 Component FAME Mixture, 47885-U Supelco). Percent fatty acid contents were calculated as mg 100g<sup>-1</sup> fish flesh by using the conversion factor presented by Greenfield and Southgate (2003) as follows:

$$FA \left( \frac{mg}{100g} \right) = \frac{[(P * FC)/100] * C * 1000}{1000} \quad (1)$$

where FA is the fatty acid content as mg per 100 grams of fish flesh; P is the fatty acid percentage; FC is the fat content of fish and C is the conversion factor (0.90 for fatty and 0.70 for lean fish).

### Statistical analyses

Statistical analyses were carried out using Microsoft Excel 2016 (Seattle, USA) software. For the comparison of the data one-way analysis of variance (ANOVA) and Tukey's multiple comparison tests were used according to species. Significance level was determined as 0.05.

### Results

Proximate compositions of horse mackerel, sole and black scorpion fish were examined in winter and summer. The moisture, ash, protein and lipid contents of the investigated species are presented in Table 1. Lipid content of horse mackerel was the highest (11.18 g 100g<sup>-1</sup>) among other investigated species and decreased to 6.31 g 100g<sup>-1</sup> in summer.

**Table 1: Proximate compositions of fish species in winter (W) and summer (S).**

g 100g <sup>-1</sup>	Horse mackerel		Sole		Black scorpion fish	
	W	S	W	S	W	S
<b>Moisture</b>	68.58 <sup>a</sup> ± 0.01	72.27 <sup>b</sup> ± 0.32	80.07 <sup>a</sup> ± 0.58	78.69 <sup>b</sup> ± 0.06	79.35 <sup>a</sup> ± 0.34	80.72 <sup>b</sup> ± 0.08
<b>Ash</b>	1.43 <sup>a</sup> ± 0.17	1.63 <sup>a</sup> ± 0.14	0.94 <sup>a</sup> ± 0.11	1.15 <sup>b</sup> ± 0.10	0.78 <sup>a</sup> ± 0.06	1.02 <sup>b</sup> ± 0.02
<b>Protein</b>	18.62 <sup>a</sup> ± 0.12	18.11 <sup>b</sup> ± 0.19	17.61 <sup>a</sup> ± 0.20	18.50 <sup>b</sup> ± 0.22	18.67 <sup>a</sup> ± 0.46	17.22 <sup>b</sup> ± 0.10
<b>Lipid</b>	11.18 <sup>a</sup> ± 0.04	6.31 <sup>b</sup> ± 0.53	0.66 <sup>a</sup> ± 0.07	0.49 <sup>b</sup> ± 0.02	0.80 <sup>a</sup> ± 0.09	0.36 <sup>b</sup> ± 0.04

<sup>a, b</sup>: Different letters show statistical significance ( $p < 0.05$ ) within the same species for winter and summer samples.

The fatty acid profiles of horse mackerel, sole and black scorpion fish in winter and summer seasons are given in Table 2. Total fatty acids of the winter samples were higher than the summer samples. Horse mackerel in winter had the highest level of total fatty acids (9834.59 mg 100g<sup>-1</sup>) among other fishes and seasons. Black scorpion fish had the second highest

fatty acid content (5444.40 mg 100g<sup>-1</sup>) followed by sole (437.05 mg 100g<sup>-1</sup>) in winter; however there was a slight difference between them. Palmitic acid (C16:0), stearic acid (C18:0), oleic acid (C18:1,  $\omega$ -9) and DHA (C22:6,  $\omega$ -3) were the fatty acids with the highest levels in all species.

**Table 2: Fatty acid profiles of horse mackerel, sole and black scorpion fish in winter (W) and summer (S).**

mg 100g <sup>-1</sup> fish flesh		Horse mackerel		Sole		Black scorpion fish	
		W	S	W	S	W	S
	<b>Σ Fatty Acids</b>	9834.59 <sup>a</sup> ±38.42	5395.77 <sup>b</sup> ±0.40	437.05 <sup>a</sup> ±1.11	333.156 <sup>b</sup> ±0.00	544.40 <sup>a</sup> ±0.28	245.61 <sup>b</sup> ±0.089
	Myristic acid C14:0	359.72 <sup>a</sup> ±0.71	278.84 <sup>b</sup> ±0.00	12.36 <sup>a</sup> ±0.03	9.24 <sup>b</sup> ±0.12	10.08 <sup>a</sup> ±0.00	6.48 <sup>b</sup> ±0.04
	Pentadecanoic acid C15:0	70.43 <sup>a</sup> ±1.42	47.70 <sup>b</sup> ±0.00	3.72 <sup>a</sup> ±0.03	1.59 <sup>b</sup> ±0.17	3.16 <sup>a</sup> ±0.12	1.15 <sup>b</sup> ±0.05
	Palmitic acid C16:0	2634.23 <sup>a</sup> ±8.54	1291.97 <sup>b</sup> ±0.80	68.03 <sup>a</sup> ±0.42	47.25 <sup>b</sup> ±0.75	95.96 <sup>a</sup> ±0.44	29.81 <sup>b</sup> ±0.36
	Heptadecanoic acid C17:0	75.46 <sup>a</sup> ±0.00	34.64 <sup>b</sup> ±0.00	8.75 <sup>a</sup> ±0.03	5.76 <sup>b</sup> ±0.15	5.35 <sup>a</sup> ±0.04	1.08 <sup>b</sup> ±0.07
	Stearic acid C18:0	1108.83 <sup>a</sup> ±5.69	366.01 <sup>a</sup> ±0.40	27.95 <sup>a</sup> ±0.13	21.27 <sup>b</sup> ±0.29	52.11 <sup>a</sup> ±0.04	13.32 <sup>b</sup> ±0.05
	Behenic acid C22:0	35.21 <sup>a</sup> ±1.42	<0.10 <sup>b</sup>	1.64 <sup>a</sup> ±0.03	1.65 <sup>a</sup> ±0.146	0.50 <sup>a</sup> ±0.00	0.28 <sup>b</sup> ±0.00
	Tricosanoic acid C23:0	19.12 <sup>a</sup> ±0.00	8.519 <sup>b</sup> ±0.00	11.06 <sup>a</sup> ±0.03	7.70 <sup>b</sup> ±0.36	7.76 <sup>a</sup> ±0.04	2.18 <sup>b</sup> ±0.089
	Lignoceric acid C24:0	38.74 <sup>a</sup> ± 0.71	33.51 <sup>b</sup> ±0.00	7.76 <sup>a</sup> ±0.00	9.76 <sup>b</sup> ±0.02	6.47 <sup>a</sup> ±0.12	2.97 <sup>b</sup> ±0.07
	<b>ΣSFA</b>	4341.75 <sup>a</sup> ±15.65	2061.19 <sup>b</sup> ±0.40	141.28 <sup>a</sup> ±0.52	104.22 <sup>b</sup> ±0.61	181.38 <sup>a</sup> ±0.63	57.27 <sup>b</sup> ±0.44
	Myristoleic acid C14:1	19.62 <sup>a</sup> ±0.71	14.20 <sup>b</sup> ±0.00	5.82 <sup>a</sup> ±0.00	4.77 <sup>b</sup> ±0.10	0.84 <sup>a</sup> ±0.08	0.49 <sup>b</sup> ±0.02
	Pentadecenoic acid C15:1	11.07 <sup>a</sup> ±0.00	<0.10 <sup>b</sup>	3.28 <sup>a</sup> ±0.06	1.75 <sup>b</sup> ±0.00	1.46 <sup>a</sup> ±0.08	0.44 <sup>b</sup> ±0.02
	Palmitoleic acid C16:1	613.78 <sup>a</sup> ±2.85	264.64 <sup>b</sup> ±0.00	42.30 <sup>a</sup> ±0.29	22.35 <sup>b</sup> ±0.22	24.81 <sup>a</sup> ±0.16	11.35 <sup>b</sup> ±0.12
	Heptadecenoic acid C17:1	45.28 <sup>a</sup> ±0.00	9.94 <sup>b</sup> ±0.40	3.19 <sup>a</sup> ±0.00	1.61 <sup>b</sup> ±0.10	3.00 <sup>a</sup> ±0.04	1.11 <sup>b</sup> ±0.07
	Oleic acid C18:1 $\omega$ -9	2340.42 <sup>a</sup> ±12.81	625.83 <sup>b</sup> ±0.00	61.35 <sup>a</sup> ±0.20	50.87 <sup>b</sup> ±0.68	102.96 <sup>a</sup> ±0.20	55.75 <sup>b</sup> ±0.41
	Gadoleic acid C20:1	123.26 <sup>a</sup> ±0.71	179.46 <sup>b</sup> ±0.00	15.73 <sup>a</sup> ±0.03	9.17 <sup>b</sup> ±0.85	8.46 <sup>a</sup> ±0.08	6.75 <sup>b</sup> ±0.46
	Erucic acid C22:1 $\omega$ -9	615.79 <sup>a</sup> ±4.27	42.02 <sup>b</sup> ±0.00	12.01 <sup>a</sup> ±0.06	9.69 <sup>b</sup> ±0.07	34.13 <sup>a</sup> ±0.12	13.56 <sup>b</sup> ±0.17
	Nervonic acid C24:1 $\omega$ -9	72.45 <sup>a</sup> ±2.85	77.23 <sup>b</sup> ±0.00	4.06 <sup>a</sup> ±0.00	2.47 <sup>b</sup> ±0.68	1.88 <sup>a</sup> ±0.12	1.21 <sup>b</sup> ±0.21
	<b>ΣMUFA</b>	3841.67 <sup>a</sup> ±15.65	1213.32 <sup>b</sup> ±0.40	147.75 <sup>a</sup> ±0.46	102.68 <sup>b</sup> ±0.80	177.52 <sup>a</sup> ±0.40	90.67 <sup>b</sup> ±0.32
	Linoleic acid C18:2 $\omega$ -6	304.38 <sup>a</sup> ±2.13	137.15 <sup>b</sup> ±0.40	19.98 <sup>a</sup> ±0.03	10.72 <sup>b</sup> ±0.36	22.37 <sup>a</sup> ±0.04	10.17 <sup>b</sup> ±0.05
	$\alpha$ -Linolenic acid C18:3 $\omega$ -3	157.97 <sup>a</sup> ±0.00	113.01 <sup>b</sup> ±0.00	5.64 <sup>a</sup> ±0.00	3.89 <sup>b</sup> ±0.02	13.38 <sup>a</sup> ±0.16	9.56 <sup>b</sup> ±0.16
	$\gamma$ -Linolenic acid C18:3 $\omega$ -6	28.68 <sup>a</sup> ±0.71	8.52 <sup>b</sup> ±0.00	3.93 <sup>a</sup> ±0.00	4.06 <sup>a</sup> ±0.12	2.10 <sup>a</sup> ±0.04	0.43 <sup>b</sup> ±0.07
	Stearidonic acid C18:4 $\omega$ -3	101.63 <sup>a</sup> ±1.42	31.80 <sup>b</sup> ±0.00	6.07 <sup>a</sup> ± 0.03	5.85 <sup>a</sup> ±0.27	3.53 <sup>a</sup> ±0.08	0.97 <sup>b</sup> ±0.05
	Eicosadienoic acid C20:2 $\omega$ -6	30.19 <sup>a</sup> ±0.00	11.07 <sup>b</sup> ±0.40	5.93 <sup>a</sup> ±0.03	2.93 <sup>b</sup> ±0.17	3.19 <sup>a</sup> ±0.08	1.60 <sup>b</sup> ±0.05
	Dihomo- $\gamma$ -Linolenic acid C20:3 $\omega$ -6	5.03 <sup>a</sup> ±0.00	<0.10 <sup>b</sup>	1.11 <sup>a</sup> ±0.13	1.423 <sup>b</sup> ±0.07	0.34 <sup>a</sup> ±0.00	0.10 <sup>b</sup> ±0.00
	Arachidonic acid C20:4 $\omega$ -6	33.71 <sup>a</sup> ±0.71	25.84 <sup>b</sup> ±0.40	4.07 <sup>a</sup> ±0.00	2.62 <sup>b</sup> ±0.02	3.02 <sup>a</sup> ±0.00	1.91 <sup>b</sup> ±0.04
	Eicosapentaenoic acid C20:5 $\omega$ -3	82.51 <sup>a</sup> ±0.00	532.12 <sup>b</sup> ±0.80	24.28 <sup>a</sup> ±0.03	25.84 <sup>b</sup> ±0.02	40.60 <sup>a</sup> ±0.07	7.80 <sup>b</sup> ±0.12
	Docosadienoic acid C22:2 $\omega$ -6	<0.10 <sup>a</sup>	<0.10 <sup>a</sup>	3.92 <sup>a</sup> ±0.00	3.50 <sup>b</sup> ±0.10	3.44 <sup>a</sup> ±0.12	<0.10 <sup>b</sup>
	Docosapentaenoic acid C22:5 $\omega$ -3	195.20 <sup>a</sup> ±1.42	87.46 <sup>b</sup> ±0.00	29.868 <sup>a</sup> ±0.03	20.39 <sup>b</sup> ±0.27	14.95 <sup>a</sup> ±0.08	8.91 <sup>b</sup> ±0.16
	Docosahexaenoic acid C22:6 $\omega$ -3	710.88 <sup>a</sup> ±4.98	1173.28 <sup>b</sup> ±1.60	43.22 <sup>a</sup> ±0.10	45.02 <sup>b</sup> ±0.65	78.57 <sup>a</sup> ±0.32	56.22 <sup>b</sup> ±0.61
	<b>ΣPUFA</b>	1650.17 <sup>a</sup> ±7.11	2120.25 <sup>b</sup> ±1.20	148.02 <sup>a</sup> ±0.13	126.26 <sup>b</sup> ±0.19	185.50 <sup>a</sup> ±0.75	97.67 <sup>b</sup> ±0.04
	<b>EPA+DHA</b>	793 <sup>a</sup>	1705 <sup>b</sup>	67 <sup>a</sup>	71 <sup>b</sup>	119 <sup>a</sup>	64 <sup>b</sup>
	<b>Σ <math>\omega</math>-3</b>	1248 <sup>a</sup>	1938 <sup>b</sup>	109 <sup>a</sup>	101 <sup>a</sup>	151 <sup>a</sup>	83 <sup>b</sup>
	<b>Σ <math>\omega</math>-6</b>	402 <sup>a</sup>	183 <sup>b</sup>	39 <sup>a</sup>	25 <sup>b</sup>	34 <sup>a</sup>	14 <sup>b</sup>
	<b><math>\omega</math>-6:<math>\omega</math>-3</b>	0.322 <sup>a</sup>	0.094 <sup>b</sup>	0.358 <sup>a</sup>	0.247 <sup>b</sup>	0.225 <sup>a</sup>	0.169 <sup>b</sup>
	Unidentified	228.41 <sup>a</sup> ±38.42	284.23 <sup>a</sup> ±0.40	24.95 <sup>a</sup> ±1.11	9.84 <sup>b</sup> ±0.00	15.58 <sup>a</sup> ±0.28	6.38 <sup>b</sup> ±0.09

a, b: Different letters show statistical difference ( $p < 0.05$ ) between winter and summer samples' fatty acids of each species.

Horse mackerel had the maximum content of PUFAs (2120.25 mg 100g<sup>-1</sup>) in summer among all species and seasons. Black scorpion fish had higher PUFA (185.50 mg 100g<sup>-1</sup>) than sole (148.02 mg 100g<sup>-1</sup>) in winter. PUFA of sole slightly changed in winter and summer, while PUFA of black scorpion fish was half that of the summer samples.

Total ω-3 content of horse mackerel in summer was 1938 mg 100g<sup>-1</sup> which was higher than in winter (1248 mg 100g<sup>-1</sup>). Dissimilarly, total ω-3 values of sole and black scorpion fish were higher in winter than in summer. Total ω-6 fatty acid contents in all species diminished in summer. The ratio of ω-6:ω-3 fatty acid of all species were between 0.094 (horse mackerel in summer) and 0.358 (sole in winter).

DHA of horse mackerel was 1173.28 mg 100g<sup>-1</sup> while EPA was 532.12 mg 100g<sup>-1</sup> in the summer samples. Horse mackerel had the highest EPA and DHA among other investigated species. The EPA level of horse mackerel was remarkably greater in summer; however there was an acute decrease in levels in black scorpion fish. On the other hand EPA and DHA levels in sole slightly changed through seasons.

### Discussion

According to Karakoltsidis *et al.* (1995) the main two components affected by the seasons were moisture and lipid. In our study, moisture contents of horse mackerel and black scorpion fish increased in summer, in contrast to sole. On the other hand, lipid contents of the investigated species were always higher

in winter. The lipid content in horse mackerel was the highest and decreased nearly to half in summer. Similar to our results, Özden (2010) stated that the fat content of horse mackerel muscle significantly decreased by summer. It was also presented that horse mackerel caught from the Black Sea had similar lipid values (Boran and Karaçam, 2011; Chuang *et al.*, 2012). In contrast with our experimental observation, lowest lipid values in winter were given by Garcia-Moreno *et al.* (2013) for horse mackerel caught from the west Mediterranean Sea. Celik (2008) also reported very low lipid contents (0.40 in winter and 2.03 in spring) from the north eastern Mediterranean Sea. In this study, horse mackerel was fatty while sole and black scorpion fish were lean fish considering the lipid composition of these species. As suggested by Ackman (1990), fish species could be categorized as lean fish (<2%); low fat (2-4%); medium fat (4-8%) and high fat (>8%) fish considering the total fat content.

It was determined that total fatty acids of the winter samples were higher than the summer samples. In our study, saturated fatty acids (SFAs) and monounsaturated fatty acids (MUFAs) of the investigated species were lower in summer as compared to winter. Considering % of fatty acids, Orban *et al.* (2011) determined that there were no significant difference between the SFAs and MUFAs of horse mackerel neither in March nor in December. Ozogul *et al.* (2011) stated that SFAs and MUFAs of sole were higher in spring than in winter.

Palmitic acid was the most abundant fatty acid found in horse mackerel both in winter and summer, and in sole only in winter. However, oleic acid was the most abundant fatty acid in sole in summer and black scorpion fish in winter. On the other hand DHA was the fatty acid with the highest level in summer in black scorpion fish. Celik (2008) and Orban *et al.* (2011) also observed that palmitic acid, stearic acid, oleic acid, and DHA were the most abundant fatty acids in horse mackerel. Gökçe *et al.* (2004) stated that DHA (in August) and palmitic acid (in February) were the highest fatty acids in sole unlike our study. Black scorpion fish caught in July to October from the Black Sea (Chuang *et al.*, 2012) and June and July from the Mediterranean Sea (Reale *et al.*, 2006) had the highest DHA and palmitic acid fatty acids. All the researchers above gave their results as percentage of total lipid.

Horse mackerel had the maximum content of PUFAs in summer. Likewise, Bandarra *et al.* (2001) reported that the maximum PUFA value of horse mackerel was in August (2356 mg 100g<sup>-1</sup>). In all investigated fish species except horse mackerel, the total amounts of PUFA were higher in winter than in summer. Among PUFAs, ALA (C18:3,  $\omega$ -3) is an essential fatty acid which cannot be synthesized by humans, and must be acquired through diet (Stark *et al.*, 2008). In the present study, winter samples had greater contents of ALA and horse mackerel was found to be the best source of this essential fatty acid. Being in the second place, black scorpion fish had higher

ALA than sole in which the amounts of ALA were much lower.

Total  $\omega$ -3 content of horse mackerel in summer was higher than in winter. It was observed that total  $\omega$ -6 fatty acid contents were lower than  $\omega$ -3 and decreased by summer in all species. In our study  $\omega$ -6: $\omega$ -3 fatty acid ratios of all species were similar to literature: 0.08 for horse mackerel (Bayır *et al.*, 2006); 0.18 for sole (Ozogul *et al.*, 2011) and 0.28 for black scorpion fish (Reale *et al.*, 2006). For a balanced intake of fatty acids the ratio is suggested to be 1:1. Nevertheless we actually consume foods with more  $\omega$ -6 in our western style diet, promoting several diseases (Simopoulos, 2002). Therefore fish is a great food source with low  $\omega$ -6 and high  $\omega$ -3 fatty acids to meet our nutritional requirements.

In the present study DHA of horse mackerel was nearly twice as that of EPA in summer. Similarly, Bayır *et al.* (2006) stated that horse mackerel had significantly higher DHA than EPA. Our results on EPA and DHA of sole were compatible with the results of İmre and Sağlık (1998) (EPA: 13 mg 100g<sup>-1</sup>; DHA: 35 mg 100g<sup>-1</sup>), however they were quite lower than the results of Candela *et al.* (1997) (EPA: 730 mg 100g<sup>-1</sup>; DHA: 590 mg 100g<sup>-1</sup>).

Horse mackerel had the highest EPA value in summer among all other species and seasons. Similar to our study, it was reported that EPA value of horse mackerel (*T. mediterraneus*) was higher in summer (713.7 mg 100g<sup>-1</sup>) (Garcia-Moreno *et al.*, 2013). In our study, DHA level of horse mackerel was considerably higher in summer.

Likewise, Garcia-Moreno *et al.* (2013) stated that DHA value of horse mackerel (*T. mediterraneus*) were higher in summer (624.5 mg 100g<sup>-1</sup>) than in winter (256.3 mg 100g<sup>-1</sup>). Also, black scorpion fish and sole had much lower DHA in comparison with that in horse mackerel.

Containing the highest amount of PUFA, horse mackerel was the fattiest fish in winter among the other investigated species. However, summer samples of horse mackerel had more EPA and DHA than winter samples. It is estimated that the difference in EPA and DHA contents in seasons is related to the change in fat content of the nourishment that fish take in summer and winter. Besides, the low amount of fat content in the summer period is due to the fact that the fat accumulation is directed to the reproductive organs depending on the spawning processes of the fish.

It is recommended to take 500 to 1800 mg EPA+DHA daily to reduce the risks relevant to mortality (Kris-Etherton *et al.*, 2002). Containing 1705 mg/100g total EPA+DHA in summer, horse mackerel samples affluently met this requirement. On the other hand, total  $\omega$ -3 contents of sole and black scorpion fish were relatively low.

DPA (C22:5,  $\omega$ -3) is another  $\omega$ -3 long chain PUFA, but it is found in small quantities in fish compared to EPA and DHA. Yet there is no pure DPA commercially available for human consumption (e.g. as supplements) (EFSA, 2012), fish itself should be consumed to benefit all long chain  $\omega$ -3 PUFA. In this study highest DPA

belonged to horse mackerel in winter. Likewise, Garcia-Moreno *et al.* (2013) stated that winter amount of DPA in horse mackerel was higher than the summer value.

In available literature, most of the research articles present their results as % of fatty acids. This way of stating the data can lead to misunderstandings from the point of consumers and scientific manner while calculating the intake values of fatty acids for a healthy diet. Consumers could only know how much of their EPA and DHA needs are met when they eat 100 grams fish by clarifying the results and reporting as mg 100g<sup>-1</sup> or g 100g<sup>-1</sup>. There were only a few studies belonged to horse mackerel and sole to compare our results with; however there were no results found regarding black scorpion fish in terms of mg 100g<sup>-1</sup>.

To conclude, horse mackerel was the best source of EPA and DHA especially in summer and completely fulfilled the daily needs of human beings. Horse mackerel in summer had 1705 mg 100g<sup>-1</sup> EPA+DHA meeting the daily need which is recommended to be 500 to 1800 mg EPA+DHA to reduce the risks relevant to mortality and support overall health. As our ancestors did forty thousand years ago, it is recommended to consume fish itself. Instead of taking only  $\omega$ -3 supplements, consuming fish provides to also benefit other long chain PUFA like ALA and DPA, protein, and other macro and micro nutrients, besides EPA and DHA fatty acids.

### Acknowledgements

This research was supported by the Research Fund of Istanbul University with the project number of BYP-29677.

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