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# Original Article

# Seasonal and sexual variations of fatty acid composition in fillet of Capoeta erhani

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**Abstract:** The lowest lipid levels of *Capoeta erhani* observed in winter and vice versa in summer. The fatty acid composition of the fillets was significantly different among seasons (*P*<0.05), while the variations among sexes were not at the same degrees (*P*>0.05). The ratios of the unsaturated fatty acids (UFAs) were higher than half of the total fatty acids among all seasons. The level of PUFA was highest in autumn (25.91%), and lowest in summer (22.11%). Among seasons and sexes, the levels of total n3 PUFAs in total fatty acids changed from 15.43% to 21.89% and n6 PUFAs from 3.8% to 7.97%, respectively. The level of n3 PUFAs was present in excess that of the n6 PUFAs. The ratios of the n3 PUFAs to n6 PUFAs in the fillets of *C. erhani* were highest in autumn for both sexes and remarkably influenced by seasons.

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#### Introduction

Oil, especially fish oil is important food source because of providing physiological and energy needs of human. Fats absorb hydrophobic vitamins and regulate cholesterol metabolism (Connor, 2000; Jabeen, 2010). The importance of fats is due to their polyunsaturated fatty acid content (Aras, 2003; Louly, 2011). Fatty acids including eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) were found mostly in fish oil contributing to the reduction of cardiovascular diseases, regulating the synthesis of prostaglandins and helping cancer prevention (Marichamy, 2012).

It is known that neurons do not have regeneration capability and neuronal membranes have little generative capabilities. PUFAs comprise 1/3 of fatty acids content of nervous system (Bourre, 1997). Therefore, fatty acids of fish oils are not only essential for normal growth and development, but also contain anti-immune system properties. In addition, they are used in production of

# pharmacological drugs.

Fishes are the important sources of proteins, vitamins; essential minerals and rich n3 long chain polyunsaturated fatty acids (Jabeen, 2010). The previous studies indicate that nutritional value, especially fatty acid composition of fish, changes with season, sex, species, salinity and temperature (Gills, 1984; Uysal, 2011). Therefore, the determination of season that fish have the best nutritional quality in terms of fatty acid composition is important for human diet.

Capoeta erhani is a member of cyprinid family and found in the Ceyhan River of eastern Anatolia. It is a benthic freshwater species. Since, there is no reports found concerning the fatty acid composition of this fish. Therefore, we aimed to determine the seasonal and sexual nutritional values and the fatty acid compositions of *C. erhani* in this study.

## Materials and Methods

Fish sampling and experimental procedures: The

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Table 1. Seasonal variation of fatty acid ratios in fillets of female *C. erhani* (% of total fatty acids).

Fatty Acids	Spring	Summer	Autumn	Winter
C12:0	$1.832 \pm 0.17$	$0.762 \pm 0.34$	$0.284 \pm 0.16$	$1.234 \pm 0.60$
C13:0	$0.102 \pm 0.05$	$0.087 \pm 0.04$	$0.07 \pm 0.02$	$0.086 \pm 0.04$
C14:0	$6.234 \pm 0.60$	$5.105 \pm 0.35$	$4.356 \pm 0.72$	$5.938 \pm 0.94$
C14:1	$0.148 \pm 0.02$	$0.167 \pm 0.04$	$0.13 \pm 0.04$	$0.142 \pm 0.02$
C15:0	$0.41 \pm 0.11$	$0.882 \pm 0.73$	$1.304 \pm 0.51$	$0.47 \pm 0.20$
C15:1	$0.062 \pm 0.02$	$0.135 \pm 0.08$	$0.182 \pm 0.06$	$0.072 \pm 0.04$
C16:0	$18.942 \pm 0.89^{a}$	$18.357 \pm 1.80^{a}$	$20.066 \pm 1.43^a$	$19.224 \pm 0.63^a$
C16:1	$8.392 \pm 2.28^a$	$11.355 \pm 3.01^{a}$	$10.922 \pm 3.60^a$	$9.45 \pm 2.37^{a}$
C17:0	$0.26 \pm 0.09$	0.37-0.14	$0.492 \pm 0.15$	$0.31 \pm 0.20$
C17:1	$0.156 \pm 0.03$	$0.255 \pm 0.12$	$0.304 \pm 0.10$	$0.164 \pm 0.06$
C18:0	$1.658 \pm 0.39$	$1.852 \pm 0.34$	$3.626 \pm 1.00$	$1.72 \pm 0.37$
C18:1n9	$21.464 \pm 2.16^{b}$	$14.867 \pm 4.71^{a}$	$10.734 \pm 4.73^{a}$	$20.124 \pm 5.16^b$
C18:1n7	$2.836 \pm 0.78$	$3.03 \pm 0.38$	$2.798 \pm 0.79$	$2.888 \pm 0.88$
C18:2n6	$5.85 \pm 1.19$	$3.415 \pm 1.31$	$1.328\pm0.23$	$5.64 \pm 1.97$
C18:3n3	$0.69 \pm 0.22$	$1.137 \pm 0.61$	$1,228 \pm 0.42$	$0.602 \pm 0.28$
C20:0	$0.126 \pm 0.04$	$0.13 \pm 0.01$	$0.134 \pm 0.02$	$0,132 \pm 0.02$
C20:1	$1.638 \pm 0.13$	$1.422 \pm 0.53$	$1.058 \pm 0.41$	$1.716 \pm 0.21$
C20:3n6	$0.196 \pm 0.05$	$0.175 \pm 0.0$	$0.116 \pm 0.05$	$0.192 \pm 0.06$
C20:4n6	$0.072\pm0.02$	$0.07 \pm 0.02$	$0.13 \pm 0.02$	$0.06\pm0.02$
C20:2n6	$0.834 \pm 0.23$	$1.3 \pm 0.39$	$2.226 \pm 1.14$	$0.924 \pm 0.54$
C20:5n3	$5.23 \pm 0.73^{a}$	$7.187 \pm 1.50^{ab}$	$9.172 \pm 1.33^{b}$	$5.626 \pm 1.18^{a}$
C22:1n9	$0.066 \pm 0.02$	$0.147 \pm 0.09$	$0.15 \pm 0.03$	$0.08 \pm 0.01$
C23:0	$0.184 \pm 0.03$	$0.24 \pm 0.05$	ND	$0.196 \pm 0.03$
C22:6n3	$9.518 \pm 1.81^{a}$	$10.515 \pm 3.26^a$	$10.568 \pm 2.66^a$	$10.122 \pm 1.48^{\rm a}$
Unidentified	13.1	17.038	18.622	12.888

<sup>\*</sup> Averages followed by the same letter show no statistical differences (P>0:05). ND: Not Detected

specimens of *C. erhani* were caught from the Menzelet Dam Lake (37°41'12"N; 36°50'11"E). After biometric data analyses (i.e., weight and length), the specimens were transferred on ice from field to the laboratory. A total of 40 specimens including 10 specimens in each season (5 males and 5 females) were examined. The dorsal muscle of specimens from each sex were excised and homogenized in a blender for 3 min for subsequent lipid extraction.

*Measurements:* Total lipid extraction was performed according to the procedure of Bligh and Dyer method (1959). Methyl esters were prepared by transmethylation using 2M KOH in methanol and hexane according to Ichihara et al. (1996) with minor modifications. Extracted lipids (10 mg) were dissolved in 2 ml hexane followed by 4 ml of 2 M

methanolic KOH. The tube was then vortexed for 2 min at room temperature. After centrifugation at 4000 rpm for 10 min, the hexane layer was taken for further analysis in GC analyses.

Gas chromatographic conditions: The fatty acid composition was analyzed by GC with auto sampler (Thermo focus) equipped with a flame ionization detector and a fused silica capillary column (30 m x 0.32 mm, ID x 0.25 μm film). The oven temperature was  $140^{\circ}$ C, held for 5 min, raised to  $200^{\circ}$ C at rate of  $4^{\circ}$ C/min and to  $220^{\circ}$ C at a rate of  $1^{\circ}$ C/min, while the injector and the detector temperature were set at  $220^{\circ}$ C and  $280^{\circ}$ C, respectively. The sample size was 5 μl and the carrier gas was controlled at 16 psi. The split used was 1:40. Fatty acids were identified by comparing the retention times of FAME mixture (SUPELCO). The results were expressed in GC area

Table 1. Seasonal variation of fatty acid ratios in fillets of male *C. erhani* (% of total fatty acids).

Fatty Acids	Spring	Summer	Autumn	Winter
C12:0	$1.172 \pm 0.51$	$0.756 \pm 0.51$	$0.366 \pm 0.14$	$0.844 \pm 0.07$
C13:0	$0.07 \pm 0.01$	$0.122 \pm 0.05$	$0.067 \pm 0.01$	$0.05 \pm 0.01$
C14:0	$5.48 \pm 0.48$	$5.48 \pm 0.64$	$4.254 \pm 0.70$	$5.606 \pm 0.70$
C14:1	$0.164 \pm 0.02$	$0.168 \pm 0.03$	$0.128 \pm 0.01$	$0.142 \pm 0.04$
C15:0	$0.444 \pm 0.06$	$1.14 \pm 0.58$	$1 \pm 0.51$	$0.332 \pm 0.08$
C15:1	$0.076 \pm 0.02$	$0.162 \pm 0.07$	$0.138 \pm 0.09$	$0.046 \pm 0.01$
C16:0	$18.478 \pm 0.64^{a}$	$18.906 \pm 0.94^{a}$	$19.388 \pm 1.02^{a}$	$20.134 \pm 1.00^{a}$
C16:1	$9.4\pm1.22^{ab}$	$13.182 \pm 1.61^{b}$	$10.91 \pm 2.29^{b}$	$7.632 \pm 1.08^a$
C17:0	$0.298 \pm 0.06$	$0.488 \pm 0.17$	$0.458 \pm 0.11$	$0.202 \pm 0.07$
C17:1	$0.18 \pm 0.04$	$0.286 \pm 0.10$	$0.264 \pm 0.10$	$0.2 \pm 0.15$
C18:0	$1.82 \pm 0.25$	$2.026 \pm 0.49$	$2.502 \pm 0.66$	$1.742 \pm 0.42$
C18:1n9	$20.242 \pm 0.96^b$	$12.314 \pm 4.25^{a}$	$11.878 \pm 3.08^{a}$	$23.128 \pm 3.44^{b}$
C18:1n7	$2.988 \pm 0.39$	$3.312 \pm 0.30$	$2.832 \pm 0.54$	$2.272 \pm 0.54$
C18:2n6	$5.43 \pm 0.97$	$2.718\pm1.32$	$1.632 \pm 0.36$	$6.822 \pm 1.59$
C18:3n3	$0.726 \pm 0.09$	$1.172\pm0.47$	$1.126\pm0.36$	$0.522 \pm 0.13$
C20:0	$0.134 \pm 0.01$	$0.194 \pm 0.10$	$0.128 \pm 0.01$	$0.114 \pm 0.02$
C20:1	$1.916 \pm 0.09$	$1.308\pm0.32$	$1.174\pm0.15$	$1.884 \pm 0.15$
C20:3n6	$0.212 \pm 0.05$	$0.154 \pm 0.03$	$0.128 \pm 0.02$	$0.202 \pm 0.02$
C20:4n6	$0.068 \pm 0.02$	$0.102\pm0.04$	$0.108 \pm 0.04$	$0.08 \pm 0.01$
C20:2n6	$0.936 \pm 0.22$	$1.79 \pm 0.66$	$2.152 \pm 0.76$	$0.872 \pm 0.33$
C20:5n3	$5.452 \pm 0.28^a$	$7.182 \pm 1.36^{b}$	$9.186 \pm 0.87^{c}$	$5.056\pm0.78^a$
C22:1n9	$0.076 \pm 0.01$	$0.136 \pm 0.05$	$0.13 \pm 0.08$	$0.075 \pm 0.01$
C23:0	$0.197 \pm 0.07$	$0.207\pm0.02$	$0.305\pm0.03$	$0.16 \pm 0.05$
C22:6n3	$10.372 \pm 1.13^{a}$	$9\pm1.74^a$	$11.58 \pm 3.11^{a}$	$11.602 \pm 2.76^{a}$
Unidentified Fatty	13.669	17.695	18.166	10.281

<sup>\*</sup> Averages followed by the same letter show no statistical differences (P>0:05). ND: Not Detected

as percentage value.

Statistical analyses: The results were presented as means ± SD (Standard Deviation). The data was analyzed by the SPSS Package Programme, version 12. The seasonal variations and fatty acid profiles of specimens were compered using One-Way ANOVA followed by Tukey's multiple comparison tests. The sexual comparison was performed by student t-Test. The results were found significant at *P*<0.05.

### Results and Discussion

The mean weights and lengths of the collected fishes were 168.76 g and 24.92 cm, respectively. The total lipid of the *C. erhani* ranged 64.62% to 68.45%. The lipid content of *C. erhani* was maximum in summer and minimum at winter. High lipid level in summer and low lipid level in winter have noted in most

fishes (Shirai et al., 2002; Zlatanos, 2007). Decreases fillet lipid content can be as result of feeding situation. Similar and contradictory results have been reported for *Solea solea* (Gökçe, 2004), *Sander lucioperca* (Uysal, 2005), and *Cyprinus carpio* (Güler, 2008).

The fatty acid composition of *C. erhani* in both sexes was presented in Tables 1 and 2. Mean of 24 fatty acids from C12:0 to C22:6n3 were identified. Collectively, the 24 fatty acids were comprised about average 87% of the total fatty acids. There were significant differences (*P*<0.05) among seasons in the fatty acid concentrations. The fatty acids occurring in the highest proportions were palmitic (16:0), palmitoleic (16:1), oleic (18:1n9), eicosapentaenoic (EPA) (20:5n3), docosahexaenoic (DHA) (22:6n3) acids in all seasons. In particular,

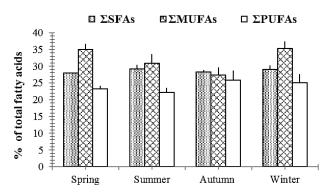


Figure 1. Seasonal variations of main fatty acid groups in fillets of male *C. erhani*.

C. erhani has more C16:0 and C18:1n9 than other fatty acids through study period. The highest concentration of Saturated Fatty Acids (SFAs) was found in summer and autumn, mainly in the form of palmitic acid (16:0). Similar results were reported by Aggelousis and Lazos (1991) and Rasoarahona et al. (2005) who noted that palmitic acid was in the highest level in autumn for three tilapia species. High values of SFAs in summer and its low levels in winter could be due to adaptation of fish to cold temperature. Logue (2000) reported that adaptation of fish species to cold water temperature is associated with increasing unsaturation. However, Bulut et al. (2012) indicated that SFAs are not affected by seasonal variations. In this study, the ratios of SFAs of male reached to the highest level in summer and that of female in autumn, but in both sexes there were not significantly change (P>0.05). Huynh (2007) noted that in both spawning and nonspawning periods, fatty acid contents decrease in the order of MUFA>SFA>PUFA. Also, Kozlova and Klotimchenko (2000) indicated that this situation was characteristic for fatty fishes. Similar result was found in present study for spring, summer and winter seasons that can be as a result of feeding period of C. erhani.

The results also showed that the major MUFA found in all samples is oleic acid (C18:1). Many studies have reported similar finding in different species (Huynh, 2007, Prato, 2012; Usydus, 2012). Oleic acid (18:1n9) reached the highest level in female and male fillets in spring and winter, respectively (Tables 1 and 2). Changes of oleic acid among seasons were

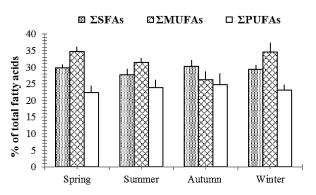


Figure 2. Seasonal variations of main fatty acid groups in fillets of female *C. erhani.* 

significant (P<0.05), but sexual variations did not show the same result (P>0.05). Usydus (2012) noted that the percentages of MUFA in Baltic sprat (Sprattus sprattus balticus) were higher during the spawning season than the feeding season. The results showed that total MUFA contents and oleic acid were similar in spring and autumn. The oleic acid has drastic roles in human diet. Lemaitre et al. (2006) reported that trans oleic acid reduces the risk of coronary heart disease. The palmitoleic acid was the second most abundant MUFA ranged from 7.63% to 13.18% in male and from 8.39% to 11.35% in female with the highest values in summer and lowest in winter for male (Tables 1 and 2). Seasonal variations in the levels of palmitoleic acid were significant in male of *C. erhani* (*P*<0.05).

The levels of PUFAs in total fatty acids were found in the range of 22.11% to 25.91% in males and 22.39% to 24.76% in females during all seasons (Figs. 1 and 2).

Eicosapentaenoic (EPA) and docosahexaenoic (DHA) acids comprise a great part of PUFAs in fillets of *C. erhani*. The levels of EPA in the fillets of *C. erhani* varied from 5.23% (spring) to 9.17% (autumn) for female; 5.05% (winter) to 9.18% (autumn) for male, and of the DHA from 9.51% (spring) to 10.56% (autumn) for female; 9% (summer) to 11.60% (winter) for male. In the light of these values, the high levels of EPA and DHA is observed in autumn and winter seasons. Therefore, it is concluded that *C. erhani* are more beneficial for human nutrition in those seasons. Çelik et al. (2005) reported that EPA contents of fishes in cold regions

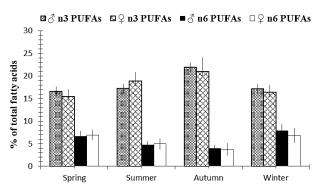


Figure 3. Seasonal and sexual variations of total n3 and n6 PUFAs in fillets of *C. erhani*.

are higher than those of warm regions. The results of present study also support this finding. The levels of EPA and DHA were influenced by seasons. But, only variations in the level of EPA were significant (P<0.05). The levels of EPA and DHA between sexes were not different remarkably. Usydus (2012) found similar results about EPA and DHA in feeding and wintering seasons. However the results on the seasonal variations of EPA and DHA were in a contrary with previous reports carried out on seasonal variations of fatty acid compositions in different species (Gökçe, 2004; Shirai et al., 2002). In addition, the percentage of DHA always exceeds that of EPA. The percentages of these n3 PUFAs in fillets depend on diet (Sargent et al., 2002) and their variations might be related to changes in the nutritional habits of the fishes (Norrobin et al., 1990).

The results showed that the levels of n3 PUFAs ranged from 15.43% to 20.96% in female and 16.55% to 21.89% in male. The levels of n6 PUFAs ranged from 3.8% to 6.95% in female and 4.02% to 7.97% in male (Fig. 3) indicating that the levels of n3 PUFAs reach the highest levels in autumn in both sexes. But the opposite situation was observed in n6 PUFAs levels especially in female. The levels of n3 PUFAs exceeded that of the n6 PUFAs. Similar results were observed by Huynh (2007), Mnari (2006) and Zlatanos (2007).

The n3/n6 ratio is a proper indicator of the nutritional value of fishes (Piggott and Tucker, 1990). The high level of n3/n6 fatty acid ratio is essential in the diet of human beings to decrease plasma lipids and

reducing heart disease and cancer risk (Kinsella et al., 1990). The results of the present study showed that the n3/n6 ratios of *C. erhani* were relatively high. The n3/n6 ratios were highest (approximately 5.5) in autumn in both male and female. Considering the n3/n6 ratios and high level of n3 PUFAs, this study revealed that *C. erhani* is a healthy food for human diet especially during autumn period.

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