



# Düzce Üniversitesi Bilim ve Teknoloji Dergisi

Research Article

## Seasonal Variations in Fat and Fatty Acid Profiles of *Barbus tauricus* (Kessler, 1877) From Duzce-Melen Basin (Turkey)

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### ABSTRACT

In this study, the total lipid and fatty acid profiles of *Barbus tauricus* samples obtained from the Melen River Basin during the winter, spring, and summer seasons were evaluated. Total lipid levels were determined at 2.27%, 2.37%, and 4.34% in winter, spring, and summer, respectively. The study also determined that the carbon count of 30 fatty acids, composing total fat compositions of *B. tauricus* caught in Melen Basin, ranges from 12 to 24. The major fatty acids of *B. tauricus* are palmitic acid and stearic acid from SFA; palmitoleic acid, trans oleic acid, oleic acid and erucic acid from MUFA and linoleic acid, alfa linolenic acid, eicosapentaenoic acid and docosahexaenoic acid from PUFA. The study is the first one on the lipid and fatty acid profiles of *B. tauricus*, one of the species living in the Duzce Melen Basin and having high economic importance, and aimed to investigate total lipid and fatty acid profiles according to the season.

**Keywords:** *Barbus tauricus*, Melen River Basin, Fatty Acids, MUFA, PUFA, SFA

## Düzce Melen Havzası (Türkiye) *Barbus tauricus* (Kessler, 1877) Populasyonu'nun Yağ ve Yağ asidi Profiline Mevsimsel Değişimi

### Öz

Bu çalışmada Melen Çayı Havzası'ndan kış, ilkbahar ve yaz mevsimlerinde alınan *Barbus tauricus* örneklerinin toplam lipid ve yağ asidi profilleri değerlendirilmiştir. Toplam lipid düzeyleri kış, ilkbahar ve yaz aylarında sırasıyla %2.27, %2.37 ve %4.34 olarak belirlenmiştir. Çalışmada ayrıca Melen Havzası'ndan avlanan *B. tauricus*'un yağ asitleri kompozisyonunda bulunan 30 yağ asidinin karbon sayılarının 12-24 arasında değiştiği tespit edilmiştir. *B. tauricus*'un başlıca yağ asitleri, SFA için palmitik asit ve stearik asit; MUFA için palmitoleik asit, trans oleik asit, oleik asit ve erusik asit ve PUFA için linoleik asit, alfa linolenik asit, eikosapentaenoik asit ve dokosaheksaenoik asittir. Bu çalışma, Düzce Melen Havzası'nda yaşayan ve ekonomik önemi yüksek türlerden *B. tauricus*'un lipid ve yağ asidi profilleri üzerine yapılan ilk çalışma olup, toplam lipid ve yağ asidi profillerini mevsimlere göre araştırmayı amaçlamaktadır.

**Anahtar Kelimeler:** *Barbus tauricus*, Melen Çayı Havzası, Yağ asitleri, MUFA, PUFA, SFA

## **I. INTRODUCTION**

Today, fish is considered a vital nutrient regarding human health unarguably. The techniques developed in recent studies can show the benefits of fat and fatty acids in fish meat on human metabolism and the species- and environment-related factors affecting the changes in their amounts. The studies on the effects of fatty acids on human health found that fatty acids are very effective in protecting against depression, heart attack, cardiovascular diseases, headaches such as migraine, high cholesterol and tension, articular rheumatism, diabetes, cancer, and some types of allergies [1], [2], [3]. In addition, rather than fish lipids' direct healing effect on diseases, especially cancer, their painkilling and protecting effects are widely accepted. Omega-3 fatty acids have essential roles in human metabolism; they accumulate in the eyes and brain in the placenta and testicles in the human body, help the functioning of these organs, and regulate lipid concentration in blood [4]. It is found that omega-3 fatty acids increase HDL levels and decrease cholesterol, triglyceride, and LDL-cholesterol levels [5].

Omega-6 fatty acids, an important member of unsaturated fatty acids, have significant roles in balancing the metabolism of eicosanoid hormones (prostaglandins, thromboxanes, and leukotrienes) which have huge effects on the human body [6]. According to the literature, omega-6 (n-6) fatty acids protect skin health by ensuring flexible and smooth skin and regulating loss of water and body temperature [7]. Studies have found that omega-3 and omega-6 fatty acids are effective in reducing the risks of cardiovascular diseases and heart attack, used in treating breast cancer. In addition, prostate cancers, and immune system diseases, have an active role in improving vision and in the development of babies' brains, preventing hypertension, allergy, immune system, and neurological system disorders [8], [9], [10], [11].

The fatty acids, including a methyl group on one end, a long hydrocarbon chain, and a carboxyl group on the other, comprise many lipids' essential elements [12]. Fatty acids are divided into saturated fatty acids (SFA) and unsaturated fatty acids. Unsaturated fatty acids are divided into monounsaturated fatty acids (MUFA) and polyunsaturated fatty acids (PUFA). PUFAs are divided into omega-3 (n-3) and omega-6 (n-6) fatty acids. The most common n-3 fatty acids in fish metabolism are  $\alpha$ -linolenic acid (ALA), docosahexaenoic acid (DPA), eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA). The fatty acids having double bonds of more than 4 and carbon atoms of more than 20 are called as HUFA (highly unsaturated fatty acids) [13], [14], [15].

Oleic acid (C18:1), the most common fatty acid in nature, comprises more than half of the fatty acids in many fats. Another fatty acid being vastly in many fats is palmitic acid (C16:0), which is a SFA. Palmitoleic acid (C16:1) and oleic acid are two important members of MUFA group. Palmitoleic acid rather exists in marine creatures, while oleic acid exists in all known natural fats [16]. The fatty acids existing in fish metabolism are palmitic (C16:0), palmitoleic (C16:1), oleic (C18:1), and eicosapentaenoic (C:20-5 n-3) and docosahexaenoic (C22:6 n-3) acids. According to carbon count and double bonds, a few fatty acids are in order, including significant linoleic (18:2 n-6) and arachidonic (20:4 n-6) acids.

Researchers reported many differences between sea and freshwater fishes in terms of fatty acids profiles, and it is found that the rate of n-6 fatty acids in freshwater fish is higher than in sea fish [17]. In freshwater fish, n-6/n-3 rates are 0.37 on average, while it is about 0.16 in sea fish. This difference is because fishes need PUFAs more for physiological adaptation to different habitats or nutrition regimes [18]. On the other hand, lipids are one of the most important energy sources of animal organisms; they have important roles as the source of fat-soluble vitamins and fatty acids. They involve creating the main elements of the cell membrane. Also, the raw materials for prostaglandins with hormone activity are unsaturated fatty acids with long chains existing in fishes.

There are many streams fed by springs in the mountains around Duzce, where is one of the most fertile plains in the West Black Sea Region. This makes the region a critical water basin. The most important

streams in Buyuk Melen Basin are Kucuk Melen Creek, Buyuk Melen Creek, Aksu, Asar, and Ugur River, and little streams feeding them (Figure 1).

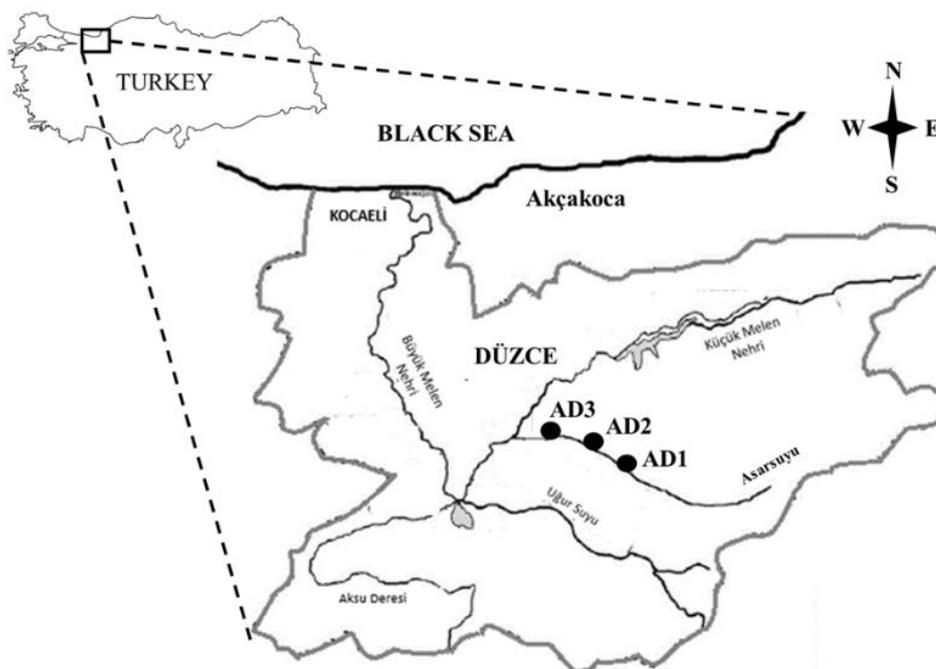
*B. tauricus* (Kessler, 1877), in the question of the study, is a species delicious and preferred in the region, which is an enormous member of the Cyprinidae family, Cypriniformes order, Actinopterygii class. Its back is darker olive green, or dark brown, sides and abdomen are yellow-brown. They generally browse around separated during spawning periods and in a large group during feeding. Apart from the spawning period, the shoaling behaviour is caused by reducing water temperatures and the gregariousness instinct [19]. The spawning period does not change according to species, but generally, it starts in May and ends in the middle of July [19].

The study aims to investigate the seasonal change of lipid and fat composition of *B. tauricus*, one of the native species living in the Duzce Melen Basin and having high commercial value. The first study, investigating the lipid and fatty acid composition of the Melen Basin population of *B. tauricus*, focuses on revealing the species' nutritional value and its seasonal lipid composition change.

## **II. MATERIAL AND METHODS**

This study was conducted at Duzce University, Faculty of Arts and Science, Biology Department, Marine Biology Research Laboratory. In addition, lipid and fatty acid analyses were done in the Seafood Processing Technology Laboratory of the Faculty of Fisheries of Mersin University.

A total of 41 *B. tauricus* samples used in the study were taken from three sampling points in the Melen Basin (Figure 1.) by buying from fishers just after the fish was caught or by the electro-fishing method. (Table 1). After the catching, samples were kept in an icebox under +4°C and transported to the laboratory as soon as possible. Sampling Data is provided in Table 1, and a map of sampling points is provided in Figure 1.



*Figure 1. Sampling area maps of B. tauricus on the Melen River Basin.*

**Table 1.** Sampling data of *B. tauricus*. Standard deviations of mean standard length (STL) and weight of samples are given brackets.

Seasons	Sampling area code	Locations	Collection Time	Sample size	Mean STL (cm) (±SD)	Mean weight (±SD)
Spring	AD1	40° 47' 44.90" N, 31° 14' 14.02" E	23.04.2018	5	13.5 (2.58)	44.08 (20.26)
	AD2	40° 49' 19.70" N, 31° 11' 43.68" E	23.04.2018	8		
Summer	AD1	40° 47' 44.90" N, 31° 14' 14.02" E	26.07.2017	9	10.54 (0.83)	21.98 (5.33)
	AD3	40° 50' 06.67" N, 31° 08' 06.11" E	26.07.2017	14		
Winter	AD2	40° 49' 19.70" N, 31° 11' 43.68" E	15.02.2018	5	13.76 (1.36)	45.37 (13.21)

## A. FAT AND FATTY ACIDS ANALYSES

Lipid content was measured by the method of Bligh and Dyer [20]. In extracted lipids, fatty acid methyl esters were obtained using the Ichibara et al. [21] method. The fatty acid composition was analyzed using a Gas Chromatography (GC) Clarus 500 device (Perkin–Elmer, USA), one flame ionization detector (FID), and SGE (60 m x 0.32 mm ID BPX70 x 0.25 µm, USA or Australia) column. Injector and detector temperatures were set as 260°C and 230°C, respectively. During this time, the furnace temperature was kept at 140 °C for 8 minutes. After that, it was increased by 4°C per minute until 220 °C, and from 220°C to 230°C by increasing the temperature 1°C per minute. It was kept at 230°C for 15 minutes to complete the analysis. The sample scale was 1 µl, and carrier gas was controlled at 16 ps. For split-flow 40.0 mL/minute (1:40) level was used. Fatty acids were determined using a comparison to the exit times of the FAME mix that contains 37 standard components.

## B. CONVERSION FACTOR

Triplicate GC analyses were performed, and the results were converted to mg fatty acid per 100 g total lipid using lipid conversion factors and then to mg fatty acid per 100 g edible portion of food using the total lipid content. Details of the derivation of lipid conversion factors were published by Weihrauch et al. [22].

$$\text{Factor (Fish)} = 0.956 - 0.143 / \text{total lipid}$$

$$\text{Fatty acid (mg/100g)} = \text{Factor} \times \text{FAME (\%)} \times \text{lipid level (\%)} \times 10$$

## C. ATHEROGENICITY INDEX (AI) AND THROMBOGENICITY INDEX (TI)

The AI and TI linked to the fatty acid composition were calculated according to Ulbricht and Southgate [23].

$$\text{AI} = [(a \cdot 12:0) + (b \cdot 14:0) + (c \cdot 16:0)] / [d \cdot (\text{PUFA } n-6+n-3) + e \cdot (\text{MUFA}) + f \cdot (\text{MUFA-18:1})]$$

$$\text{TI} = [g \cdot (14:0+16:0+(18:0))] / [(h \cdot \text{MUFA}) + i \cdot (\text{MUFA-18:1}) + (m \cdot n-6) + (n \cdot n-3) + (n-3/n-6)]$$

$$a, c, d, e, f=1, b=4, g=1, h, i, m=0.5 \quad n=3$$

## D. STATISTICAL ANALYSIS

The data obtained from the analyses of the study was evaluated by using the IBM SPSS version 22 statistics program. For evaluating lipid and fatty acids data, before statistics analysis, separation control (based on Z value) and variance homogeneity test (Duncan's Multiple Range Test) were done on all samples. The difference between groups was determined with the "one-way analysis of variance" (one-way Anova).

### III. RESULTS

#### A. TOTAL LIPID (%)

Fishes were defined into four groups non-fatty fishes (<2%), low fatty fishes (2-4%), mid-fatty fishes (4-8%), high fatty fishes (>8%) according to their fat ingredients based on Lambertsen standards, as well as into groups as non-fatty if <2%, low fatty if between 2-7%, fatty if 7-15% and high fatty if >15% according to their fat levels based on Polish Standard PN-A-86770,1999. Lipid level change of *B. tauricus* ranges in winter, spring, and summer seasons as 2.27%, 2.37%, and 4.34%, respectively; therefore, this species may be classified as a low fatty fish according to both Lambertsen and Polish standards.

#### B. FATTY ACIDS (%)

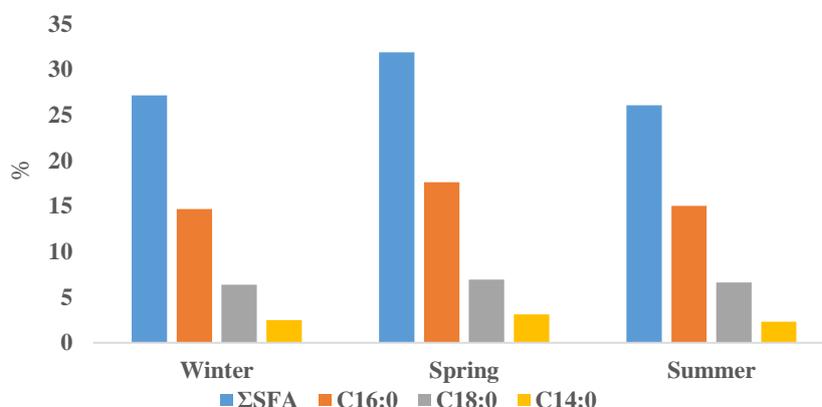
It was found that the carbon count of fatty acids in the fatty acid composition of *B. tauricus* caught from Melen Basin ranged between 12-24. *B. tauricus*'s seasonal fatty acid profile (%) is shown in Table 2.

**Table 2.** Seasonal fatty acids profile of *B. tauricus* caught from Melen River Basin (%).

Fatty acids (%)	Winter	Spring	Summer
Lauric acid (C12:0)	1.40±0.52 <sup>ab</sup>	1.81±0.63 <sup>b</sup>	0.57±0.07 <sup>a</sup>
Myristic acid (C14:0)	2.48±0.56 <sup>ab</sup>	3.11±0.27 <sup>b</sup>	2.30±0.08 <sup>a</sup>
Pentadecanoic acid (C15:0)	0.27±0.01 <sup>a</sup>	0.33±0.10 <sup>a</sup>	0.29±0.02 <sup>a</sup>
Palmitic acid (C16:0)	14.68±1.36 <sup>a</sup>	17.64±5.25 <sup>a</sup>	15.05±0.43 <sup>a</sup>
Margaric acid (C17:0)	0.46±0.15 <sup>a</sup>	0.49±0.15 <sup>a</sup>	0.37±0.02 <sup>a</sup>
Stearic acid (C18:0)	6.37±0.42 <sup>a</sup>	6.93±1.59 <sup>a</sup>	6.61±0.35 <sup>a</sup>
Arachidic acid (C20:0)	0.52±0.01 <sup>b</sup>	0.61±0.05 <sup>b</sup>	0.25±0.10 <sup>a</sup>
Behenic acid (C22:0)	0.37±0.01 <sup>a</sup>	0.37±0.06 <sup>a</sup>	0.35±0.17 <sup>a</sup>
Lignosuric acid (C24:0)	0.61±0.05 <sup>b</sup>	0.61±0.13 <sup>b</sup>	0.30±0.01 <sup>a</sup>
<b>ΣSFA</b>	<b>27.16</b>	<b>31.90</b>	<b>26.09</b>
Myristoleic acid (C14:1)	0.34±0.03 <sup>a</sup>	0.37±0.01 <sup>a</sup>	0.45±0.03 <sup>b</sup>
Pentadecenoic acid (C15:1)	0.29±0.02 <sup>a</sup>	0.36±0.03 <sup>b</sup>	0.27±0.03 <sup>a</sup>
Palmitoleic acid (C16:1)	6.14±0.41 <sup>a</sup>	6.73±1.09 <sup>a</sup>	8.67±0.45 <sup>b</sup>
Heptadecenoic acid (C17:1)	0.42±0.07 <sup>a</sup>	0.39±0.16 <sup>a</sup>	0.34±0.02 <sup>a</sup>
Transoleic acid (C18:1 <sup>n</sup> 9 <sup>t</sup> )	14.91±0.55 <sup>a</sup>	13.42±3.42 <sup>a</sup>	14.04±0.41 <sup>a</sup>
Oleic acid (C18:1 <sup>n</sup> 9 <sup>c</sup> )	4.03±0.42 <sup>ab</sup>	3.92±0.05 <sup>a</sup>	4.52±0.09 <sup>b</sup>
Vaccenic acid (C18:1 <sup>n</sup> 7)	0.12±0.01 <sup>a</sup>	0.11±0.01 <sup>a</sup>	0.13±0.03 <sup>a</sup>
Gadoleic acid (C20:1 <sup>n</sup> 9)	1.85±0.14 <sup>c</sup>	1.53±0.14 <sup>b</sup>	1.28±0.02 <sup>a</sup>
Erucic acid (C22:1 <sup>n</sup> 9)	5.13±0.45 <sup>b</sup>	4.35±0.67 <sup>b</sup>	3.31±0.10 <sup>a</sup>
Neuronic acid (C24:1 <sup>n</sup> 9)	0.00±0.00 <sup>a</sup>	0.08±0.04 <sup>b</sup>	0.06±0.00 <sup>b</sup>
<b>ΣMUFA</b>	<b>33.23</b>	<b>31.26</b>	<b>33.07</b>
Linolelaidic acid (C18:2 <sup>n</sup> 6 <sup>t</sup> )	0.00±0.00 <sup>a</sup>	0.13±0.03 <sup>b</sup>	0.21±0.05 <sup>c</sup>
Linoleic acid (C18:2 <sup>n</sup> 6 <sup>c</sup> )	6.68±0.55 <sup>a</sup>	6.96±1.12 <sup>a</sup>	6.97±0.06 <sup>a</sup>
Alfa linolenic acid (C18:3 <sup>n</sup> 3)	4.02±0.50 <sup>b</sup>	2.24±0.84 <sup>a</sup>	3.15±0.14 <sup>ab</sup>
Gama linolenic acid (C18:3 <sup>n</sup> 6)	0.45±0.04 <sup>b</sup>	0.36±0.05 <sup>a</sup>	0.37±0.00 <sup>ab</sup>
Eicosatrienoic acid (C20:3 <sup>n</sup> 3)	0.69±0.03 <sup>a</sup>	0.75±0.02 <sup>a</sup>	0.63±0.19 <sup>a</sup>
Dihomo gamma linolenic acid (C20:3 <sup>n</sup> 6)	0.41±0.04 <sup>a</sup>	0.39±0.07 <sup>a</sup>	0.52±0.12 <sup>a</sup>
Arachidonic acid (C20:4 <sup>n</sup> 6)	0.62±0.09 <sup>a</sup>	0.93±0.15 <sup>b</sup>	0.76±0.03 <sup>ab</sup>
Eicosapentaenoic acid (C20:5 <sup>n</sup> 3)	5.32±0.39 <sup>a</sup>	5.54±0.65 <sup>a</sup>	7.16±0.27 <sup>b</sup>
Adrenic acid (C22:4 <sup>n</sup> 6)	0.63±0.10 <sup>a</sup>	0.54±0.10 <sup>a</sup>	0.33±0.01 <sup>a</sup>
Docosahexaenoic acid (C22:6 <sup>n</sup> 3)	9.81±0.79 <sup>b</sup>	8.57±1.11 <sup>ab</sup>	7.71±0.12 <sup>a</sup>
Docosadienoic acid (C22:2 <sup>cis</sup> )	0.16±0.03 <sup>c</sup>	0.00±0.00 <sup>a</sup>	0.11±0.01 <sup>b</sup>
<b>ΣPUFA</b>	<b>28.79</b>	<b>26.41</b>	<b>27.92</b>
SFA/PUFA	0.94	1.21	0.93
Σ <sup>n</sup> 7	0.12	0.11	0.13
Σ <sup>n</sup> 6	8.79	9.31	9.16
Σ <sup>n</sup> 3	19.84	17.10	18.65
Σ <sup>n</sup> 9	25.92	23.30	23.21
<sup>n</sup> 6/ <sup>n</sup> 3	0.44	0.54	0.49
<sup>n</sup> 3/ <sup>n</sup> 6	2.26	1.84	2.04
DHA/EPA	1.84	1.55	1.08
AI	0.34	0.45	0.33
TI	0.25	0.34	0.27
<b>Unidentified</b>	<b>10.82</b>	<b>10.43</b>	<b>12.92</b>

<sup>x±s<sub>x</sub></sup>: means ± SD; the values in the same line with a different superscript letter (a-b-c) are significantly different (p<0.05)

The highest  $\Sigma$ SFA level was determined as 31.9% in spring and the lowest as 26.09% in summer. Dominant SFAs are palmitic acid, Myristic acid and stearic acid. The lowest amounts are pentadecanoic acid and behenic acid (Table 2, Figure 2).



**Figure 2.** Seasonal change of *B. tauricus*'s  $\Sigma$ SFA and dominant saturated fatty acids (%).

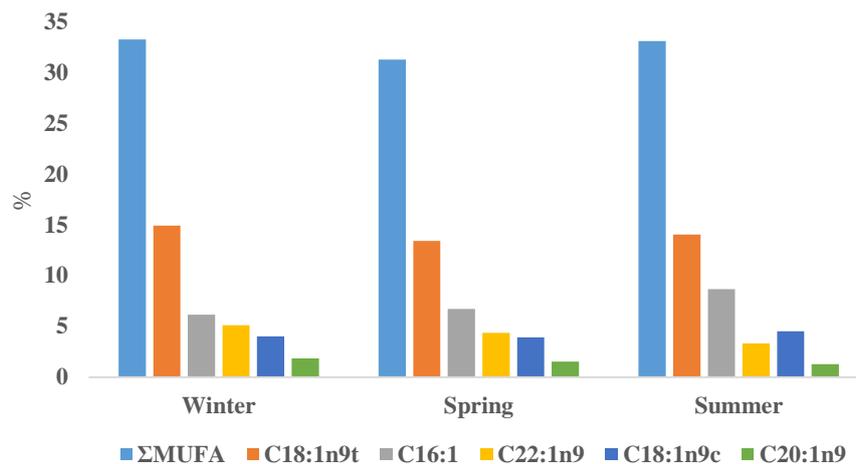
Myristic acid level is observed as 3.11% at most in spring and as 2.30% at least in summer. While there is no statistical difference in myristic acid levels between winter and summer ( $p > 0.05$ ), there is a statistical difference between these two seasons and spring ( $p < 0.05$ ). The palmitic acid level is 17.64% at most in spring and 14.68% at least in winter. No seasonal difference was observed in the palmitic acid level of *B. tauricus* ( $p > 0.05$ ). The stearic acid rate is calculated as 6.93% at most in spring and 6.37% at least in winter. Seasonally, no statistical difference in stearic acid level was observed ( $p > 0.05$ ) (Figure 2).

Lignosuric acid is observed as 0.61% at most in winter and spring and as 0.30% at least in summer. While there is no statistical difference in lingosuric acid levels between winter and spring ( $p > 0.05$ ), there is a statistical difference between these two seasons and summer ( $p < 0.05$ ). Lingosuric acid levels are very close to each other during the mid-season and remain the same during the spring and winter months. The behenic acid level of *B. tauricus* is observed as 0.37% at most in winter and spring and as 0.35% at least in summer; also, there is no statistical difference in behenic acid level between seasons ( $p > 0.05$ ) (Table 2).

$\Sigma$ MUFA is found at 33.23% at most in winter and at 31.26% at least in spring.  $\Sigma$ MUFA level demonstrates seasonal change (Table 2, Figure 3).

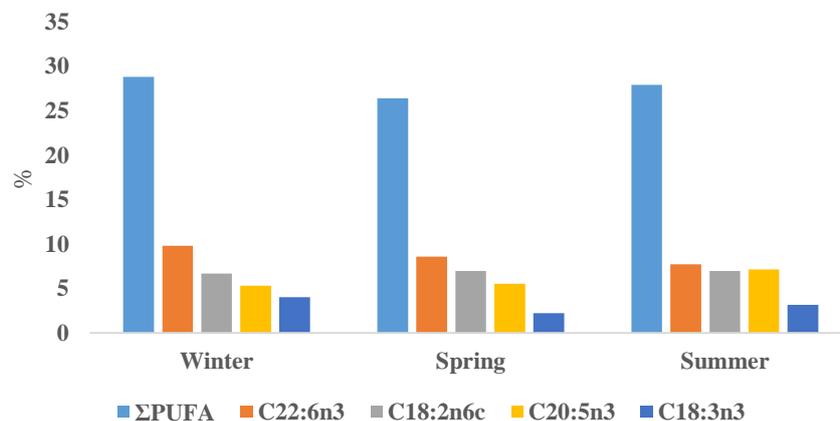
Transoleic acid is found at 14.91% at most in winter and at 13.42% at least in spring. It also is determined as the highest unsaturated fatty acid type. Palmitoleic acid is 8.67% at most in summer, 6.73% in spring, and 6.14% at least in winter. While there is no significant difference in the rate of palmitoleic acid between winter and spring ( $p > 0.05$ ), there is a statistical difference between these two seasons and summer ( $p > 0.05$ ). Heptadecanoic acid is determined as 0.42% at most in winter and 0.34% at least in summer. Oleic acid is observed as 4.52% at most in summer and as 3.92% at least in spring. Gadoleic acid is 1.85% at most in winter and 1.28% at least in summer. Erucic acid levels are calculated as 5.13% at most in winter and 3.31% at least in summer (Table 2, Figure 3).

Myristoleic acid level is 0.45% at most in summer, while 0.34% and 0.37% in winter and spring, respectively. While there is no significant difference in the rate of myristoleic acid between winter and spring ( $p > 0.05$ ), there is a statistical difference between these two seasons and summer ( $p > 0.05$ ). Pentaecenoic acid is 0.36% at most in spring and 0.27% at least in summer. Vaccenic acid is determined as 0.13% at most in summer and 0.11% at least in spring. The levels of neuronc acid, one of the monounsaturated fatty acids, is 0.08% at most in spring, while no neuronc acid is determined in winter (Table 2).



**Figure 3.** Seasonal change of *B. tauricus*'s  $\Sigma$ MUFA and dominant monounsaturated fatty acids (%).

*B. tauricus*'s  $\Sigma$ PUFA level is calculated at most in winter (28.79%) and at least in spring (26.41%). This is because levels are observed too close to each other between seasons (Table 2, Figure 4).



**Figure 4.** Seasonal change of *B. tauricus*'s  $\Sigma$ PUFA and dominant polyunsaturated fatty acids (%).

Regarding *B. tauricus*'s dominant polyunsaturated fatty acids, Docosahexaenoic acid (DHA) is observed as 9.81% at most in winter and 7.71% at least in summer. There is a statistical difference in the level of docosahexaenoic acid between winter and summer ( $p < 0.05$ ). While there is no significant difference in the rate of eicosapentaenoic acid (EPA) between winter (5.32%) and spring (5.54%) ( $p > 0.05$ ), there is a statistical difference between these two seasons and summer (7.16%) ( $p > 0.05$ ). The linoleic acid level is 6.97% at most in summer and 6.68% at least in winter; in addition, no statistical difference is observed between seasons ( $p > 0.05$ ). Alfa linolenic acid level is 4.02% at most in winter and 2.24% at least in spring. There is a statistical difference in the level of alfa linolenic acid between winter and summer ( $p < 0.05$ ) (Table 2, Figure 4).

Linolelaidic acid is 0.21% at most in summer and below the detection limit in winter. Gamma linoleic acid level is 0.45% in winter and 0.36%, and 0.37% in spring and summer. There is no statistical difference in gamma linoleic acid levels between the seasons ( $p > 0.05$ ). Eicosatrienoic acid level is 0.75% at most in spring and 0.63% at least in summer. Dihomo gamma-linolenic acid is 0.52% at most in summer and 0.39% at least in spring. Arachidonic acid is 0.93% at most in spring and 0.62% at least in winter. Adrenic acid is 0.63% at most in winter and 0.33% at least in summer, while there is no statistical difference in levels between seasons ( $p > 0.05$ ). Docosadienoic acid is not determined in spring. It is observed as 0.16% at most in winter and as 0.11% at least in summer. There is a statistical difference determined in levels of docosadienoic acid between three seasons ( $p > 0.05$ ) (Table 2).

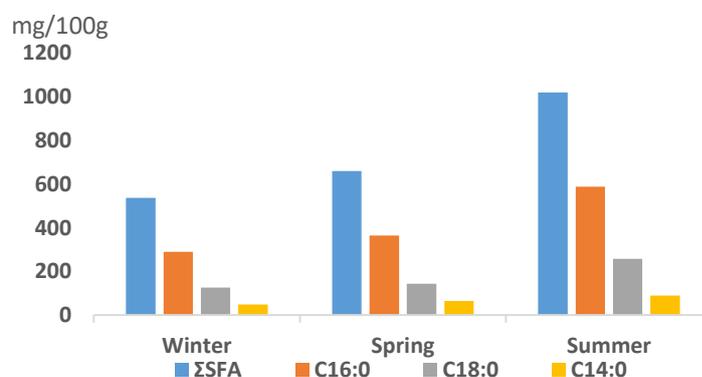
### C. FATTY ACIDS (MG/100G)

Seasonal changes in fatty acids of the species *B. tauricus* are shown in Table 3.

**Table 3.** Seasonal fatty acids profile of *B. tauricus* caught from Melen River Basin (%).

Conversion Factor	0.870	0.872	0.900
Fatty acid (mg/100g)	Winter	Spring	Summer
Lauric acid (C12:0)	27.65	37.41	22.26
Myristic acid (C14:0)	48.98	64.27	89.83
Pentadecanoic acid (C15:0)	5.33	6.82	11.33
Palmitic acid (C16:0)	289.92	364.56	587.85
Margaric acid (C17:0)	9.08	10.13	14.45
Stearic acid (C18:0)	125.80	143.22	258.19
Arachidic acid (C20:0)	10.27	12.61	9.77
Behenic acid (C22:0)	7.31	7.65	13.67
Lignosuric acid (C24:0)	12.05	12.61	11.72
<b>ΣSFA</b>	<b>536.38</b>	<b>659.26</b>	<b>1019.07</b>
Myristoleic acid (C14:1)	6.71	7.65	17.58
Pentadecenoic acid (C15:1)	5.73	7.44	10.55
Palmitoleic acid (C16:1)	121.26	139.08	338.65
Heptadecanoic acid (C17:1)	8.29	8.06	13.28
Trans oleic acid (C18:1n9t)	294.46	277.34	548.40
Oleic acid (C18:1n9c)	79.59	81.01	176.55
Vaccenic acid (C18:1n7)	2.37	2.27	5.08
Gadoleic acid (C20:1n9)	36.54	31.62	50.00
Erucic acid (C22:1n9)	101.31	89.90	129.29
Neuronic acid (C24:1n9)	0	1.65	2.34
<b>ΣMUFA</b>	<b>656.26</b>	<b>646.03</b>	<b>1291.71</b>
Linolelaidic acid (C18:2n6t)	0	2.69	8.20
Linoleic acid (C18:2n6c)	131.92	143.84	272.25
Alfa linolenic acid (C18:3n3)	79.39	46.29	123.04
Gamma linolenic acid (C18:3n6)	8.89	7.44	14.45
Eicosatrienoic acid (C20:3n3)	1.62	15.50	24.61
Dihomo gamma linolenic acid (C20:3n6)	8.10	8.06	20.31
Arachidonic acid (C20:4n6)	12.24	19.22	29.69
Eicosapentaenoic acid (C20:5n3)	105.06	114.23	279.67
Adrenic acid (C22:4n6)	12.44	11.16	12.89
Docosahexaenoic acid (C22:6n3)	193.74	177.11	301.15
Docosadienoic acid (C22:2cis)	3.16	0	4.30
<b>ΣPUFA</b>	<b>568.57</b>	<b>545.80</b>	<b>1090.55</b>
Σn6	173.59	192.40	357.79
Σn3	391.82	353.40	728.47
Σn9	511.89	481.53	906.58
Σn7	2.37	2.27	5.08
<b>Unidentified</b>	<b>213.68</b>	<b>215.55</b>	<b>504.66</b>

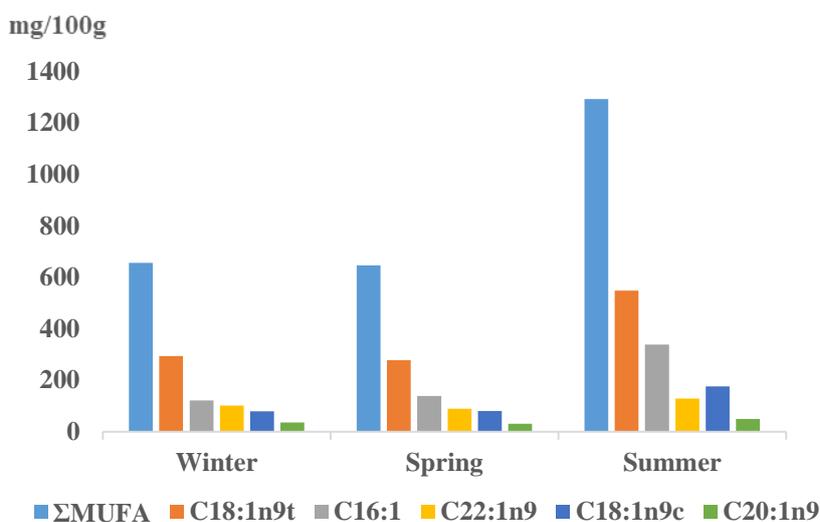
*B. tauricus*'s ΣSFA rate is 1019.07 mg/100g at most in summer and 536.38 mg/100g at least in winter (Table 3, Figure 5).



**Figure 5.** Seasonal change of *B. tauricus*'s ΣSFA and dominant saturated fatty acids (mg/100g).

The lauric acid level is 37.41 mg/100g at most in spring and 22.26 mg/100 at least in summer. Myristic acid's summer level is almost twice the time of winter level, and the level is calculated as 89.83 mg/100g in summer, 48.98 mg/100g in winter, and 64.27 mg/100g in spring. Pentadecic acid's winter and spring levels are data close, while the summer level is 11.33 mg/100g, about twice the time of the other two seasons. Palmitic acid is calculated as 587.85 mg/100g, 364.56 mg/100g, and 289.92 mg/100g in summer, winter, and spring, respectively. Margaric acid is 14.45 mg/100 at most in summer and 9.08 mg/100 g at least in winter. Stearic acid is 258.19 mg/100g at least in summer, 125.80 mg/100g at least in winter, and 143.22 mg/100g in spring. Arachidic acid is 10.27 mg/100g in winter, 9.77 mg/100g in summer, and 12.61 mg/100g at most in spring. Behenic acid is 7.31 mg/100g at least in winter. Behenic acid's summer level is 13.67 mg/100g, and the spring level is 7.65 mg/100g. Lignoceric acid level is 12.61 mg/100g at most in spring and 11.72 mg/100g at least in summer.

*B. tauricus*'s  $\Sigma$ MUFA level is 1019.07 mg/100g at most in summer and 536.38 mg/100g at least in spring (Table 3, Figure 6).

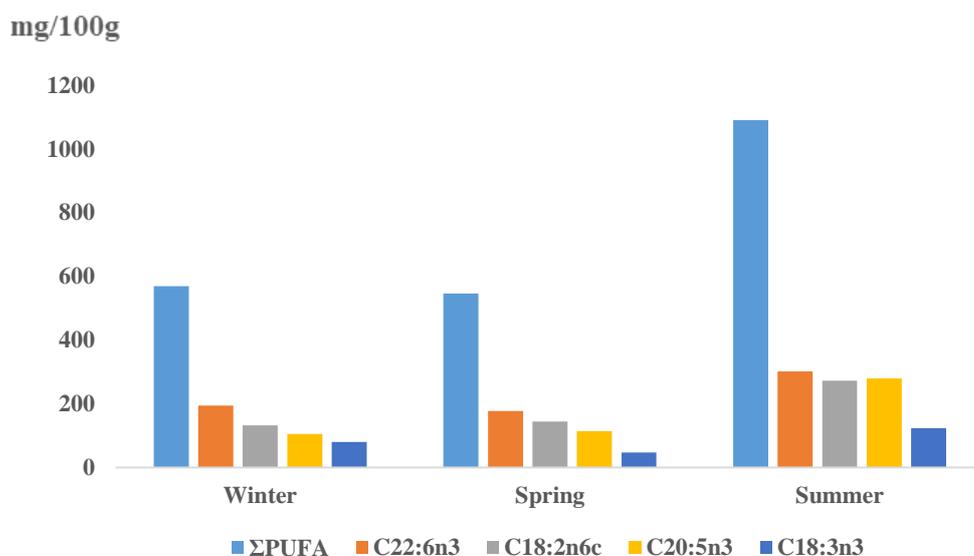


**Figure 6.** Seasonal change of *B. tauricus*'s  $\Sigma$ MUFA and dominant monounsaturated fatty acids (mg/100g).

Myristoleic acid is 17.58 mg/100g at most in summer and 6.71 mg/100g at least in winter; it also is close to the winter level with 7.65 mg/100g in spring. Pentadecenoic acid is 10.55 mg/100g at most in summer and 5.73 mg/100g at least in winter. Palmitoleic acid is 121.26 mg/100g at least in winter, 139.08 mg/100g in spring and 338.65 mg/100g at most in summer. Heptadecanoic acid is close to each other in winter and spring, with 13.28 mg/100g. In the study on the species *B. tauricus*, trans oleic acid, being a MUFA, is 548.40 mg/100g at most in summer, 277.34 mg/100g in spring, and 294.46 mg/100g at least in winter. On the other hand, oleic acid is close to each other in winter and spring and is 176.55 mg/100g at most in summer.

Vaccenic acid is 2.27 mg/100g at least in spring. Vaccenic acid's summer level is 5.08 mg/100g, and the winter level is 2.37 mg/100g. Gadaloic acid is 36.54 mg/100g in winter, 31.62 mg/100g in spring, and 50.00 mg/100g in summer. Erucic acid is 101.31 mg/100g in winter, 89.90 mg/100g in spring, and 129.29 mg/100g in summer. Neuronic acid is under the detection limit in winter and is 1.65 mg/100g in spring and 2.34 mg/100g in summer.

*B. tauricus*'s  $\Sigma$ PUFA rate is 1090.55 mg/100g at most in summer and 545.80 mg/100g at least in spring (Table 3, Figure 7).



**Figure 7.** Seasonal change of *B. tauricus*'s  $\Sigma$ PUFA and dominant polyunsaturated fatty acids (mg/100g).

Linolelaidic acid is under the detection limit in winter and is 2.69 mg/100g in spring and 8.20 mg/100g in summer. Linoleic acid is one of the highest fatty acid levels within the MUFA group and is calculated as 272.25 mg/100g in summer, 131.92 mg/100g in winter, and 143.84 mg/100g in spring. Alfa linoleic acid is 46.29 mg/100g at least in spring and 123.04 mg/100g at most in summer. Gamma linoleic acid is 14.45 mg/100g at most in summer, 7.44 mg/100g at least in spring, and 8.89 mg/100g in winter. Eicosatrienoic acid is 24.61 mg/100g at most in summer, 1.62 mg/100g at least in winter and 15.50 mg/100g in spring. Dihomo gamma-linolenic acid is 20.31 mg/100g at most in summer, while the level is lower and close to each other in winter and spring. Arachidonic acid is calculated as 12.24 mg/100g in winter, 19.22 mg/100g in spring, and 29.69 mg/100g in summer. Eicosapentaenoic acid is determined as 105.06 mg/100g in winter, 114.23 mg/100g in spring and 279.67 mg/100g in summer. Adrenic acid is 12.89 mg/100g at most in summer, 11.16 mg/100g at least in spring, and 12.44 mg/100g in winter. Docosahexaenoic acid is 301.15 mg/100g at most in summer, 177.11 mg/100g at least in spring, and 193.74 mg/100g in winter. Arachidonic acid is calculated as 12.24 mg/100g in winter, 19.22 mg/100g in spring, and 29.69 mg/100g in summer. Docosadienoic acid is determined as 4.30 mg/100g at most in summer and is under the detection limit in spring.

## **IV. DISCUSSION**

*B. tauricus*'s total lipid amounts are found at most in summer (4.34%) and at least in winter (2.27%). The reason *B. tauricus*'s total lipid amounts are low in winter is thought to be due to an increase in lipid usage by fishes needing more energy during the winter season. For poikilotherm creatures, the temperature is the most crucial factor affecting fish-feeding activity [24]. Similar results (in winter, 1.19%) were found in a study on the species *Dicentrarchus labrax* by Belikusakli [25]. Furthermore, the lipid amount of the *Squalius pursakensis* from the Melen Basin was found to be low in the winter season by Inan et al. [24]. In this study, *B. tauricus* was evaluated as a lower-fat fish based on both Lambertsen [26] and Polish (Polish Standard PNA-86770 1999) standards.

*B. tauricus*'s saturated fatty acids (SFAs) are observed as 26.09% at least in summer and 31.90% at most in spring. Unsaturated fatty acids (MUFAs) are observed as 31.26% at least in spring and as 33.23% at most in winter. Polyunsaturated fatty acids (PUFAs) are observed as 26.41% at least in spring and 28.79% at most in winter. In the present study, the MUFAs content (31.26-33.23%) was generally higher than PUFAs (26.41-28.79%) and SFAs (26.09-31.90). Similarly, MUFAs were higher in *Aspius vorax*, *Carassobarbus luteus*, *Acanthobrama marmid*, *Cyprinion macrostomum* and *Capoeta trutta* by Kacar and Bashan [27] from Ataturk Dam Lake. On the other hand, reported higher PUFA content for *Squalius*

*pursakensis* by Ates [28] from the upper Sakarya River and Inan et al. [24] from the Melen River Basin.

Researchers have found that palmitic acid is the most common fatty acid in fish tissue. In this study, palmitic acid has the highest levels among the primary saturated fatty acid in all seasons. Therefore, researchers have suggested that palmitic acid, the highest in fish's fatty acid composition, is due to its role in fatty acid metabolism [29], [30]. Similar results suggesting that palmitic acid is the essential saturated fatty acid were found by Kandemir [31] and Turchini et al. [32] for the species *Tinca tinca*, by Konar et al. [33] for the species *Capoeta trutta* and *Barbus rajanorum mystaceus*, and by Kaya and Turan [34] for anchovy.

In this study, it is found that *B. tauricus*'s  $\Sigma$ SFA (Figure 2) and  $\Sigma$ MUFA (Figure 3) rates in the Melen Basin are affected by seasonal change, while  $\Sigma$ PUFA (Figure 4) rates are close to each other between different seasons (Table 2). Fish's fatty acid compositions change based on habitat, season, nutrition, breeding, water temperature, and pollution. Melen River Basin's water amount difference is too much between seasons. And this suggests that the features of fish's habitats are constantly changing. Also, the reason for not sampling during autumn is that too much water amount not allow the sampling. This high rate of change in water amount between seasons is thought to change many factors, from water pollution to species' bio-ecology and fatty acid compositions of the fishes.

In the study, it is found that *B. tauricus*'s dominant SFAs are palmitic acid (C16:0), Myristic acid (C16:0) and stearic acid (C18:0) (Figure 2); dominant MUFAs are Transoleic acid (C18:1n9t), Palmitoleic acid (C16:1), Erucic acid (C22:1n9), Oleic acid (C18:1n9c), Gadoleic acid (C20:1n9); dominant PUFAs Docosahexaenoic acid (C22:6n3), Linoleic acid (C18:2n6c), Eicosapentaenoic acid (C20:5n3), Alfa linolenic acid (C18:3n3) (Table 2). In studies by Ozogul et al. [35], Mahmoud et al. [36], Hisar and Hisar [37] on *Cyprinus carpio*, by Inan et al. [24] on *S. pursakensis* and by Ates [28] on *Squalius pursakensis* and *Capoeta*, dominant SFAs were determined as myristic, palmitic and stearic acid, similar to our study. In studies by Gokce et al. [38] *Solea solea* has palmitic (19.00%) and stearic acid (6.91%) rates at most in summer, while in our study, palmitic (17.64%) and stearic acid (6.93%) rates were found at most in spring. The reason for this difference might be fishes living in lakes or seas and their feeding regime.

In this study, *B. tauricus*'s EPA and DHA levels were high in all seasons (Table 2). Within the fatty acid composition, DHA is highest in winter (9.81%) and lowest in summer (7.71%). On the contrary, EPA is highest in summer (7.16%) compared to other seasons. Also, it is observed that the PUFAs rate increases in winter by DHA and in summer by EPA. The reason for these changes in EPA and DHA is thought to be related to the changes in the feeding regime of *B. tauricus* during the year because EPA and DHA rates in PUFA are related to feeding [39] [40].

The n-3/n-6 ratio is a good index for comparing the relative nutritional value of fish oils [40]. The ratios obtained in this study (Table 2) suggest that n3/n6 ratios are 2.26 in winter, 1.84 in spring, and 2.04 in summer. An increase in the n-3/n-6 fatty acid ratio is essential to human nutrition in preventing coronary heart disease and reducing cancer risk [41]. Guler et al. [42] found that the n-3/n-6 fatty acids ratio is 1.49 in spring, 1.45 in autumn, and 1.22 in winter, while 0.72 at least in summer, in *Sander lucioperca*. Kalyoncu et al. [43], found that n-3 / n-6 fatty acids ratio is 1.4 in spring, 1.5 in summer, 1.2 in autumn, and 1.4 in winter, in *Vimba vimba tenella*. Satar et al. [40], found that the n-3/n-6 ratio is 3.56 in winter, 3.80 in summer, 4.94 in spring, and 3.34 in autumn in *Capoeta trutta*, and specified *C. trutta* as an important freshwater fish due to its high n-3/n-6 ratio. Considering the results of other studies on n-3/n-6 ratios of freshwater fishes investigated, *B. tauricus* can also be considered an important freshwater fish for human nutrition.

SFA/PUFA ratio (0.93-1.21) determined in this study is higher than the one obtained in studies by Inan et al. [24] (0.80-0.93) and Ates [28] (0.46-0.65) on *S. pursakensis*.

In analyzing seasonal changes in fatty acid amounts in 100gr *B. tauricus* fillet,  $\Sigma$ SFA ranges between 536.38-1019.07 mg/100g;  $\Sigma$ MUFA ranges between 646.03-1291.71 mg/100g; and  $\Sigma$ PUFA ranges

between 545.80-1090.55 mg/100g. The results ( $\Sigma$ SFA:455.25-1461.20/  $\Sigma$ MUFA:460.16-1396.64/  $\Sigma$ PUFA:492.10-1820.0) reported by Inan et al. [24] for *S. pursakensis* support our findings.

*B. tauricus* EPA amount is 105.06 mg/100g in winter, 114.23 mg/100g in spring, and 279.67 mg/100g in summer, DHA is 193.74 mg/100g in winter, 177.11 mg/100g in spring, and 301.15 mg/100g in summer. Inan et al. [24] have found EPA for *S. pursakensis* between 74.01-231.14 mg/100g and DHA between 152.16-764.06 mg/100g. Kminkova et al. [44] have found EPA for carp between 82.2-149.2 mg/100g and DHA between 24.5-56.2 mg/100g.

Fish oil is essential for human health [45]. Fish oils have roles in treating many diseases causing death, primarily cardiovascular diseases. Especially in some of the developed countries, fish have become crucial in preventing and treating cardiovascular diseases that account for 40% of deaths [46]. This study and similar studies show that fish-originated nutrition should be encouraged for human health [47].

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