

Seasonal changes in the fatty acid profiles of widely-consumed fish species from the Mesopotamian River Basin

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SUMMARY: This study investigates seasonal variations in total lipid content and fatty acid composition of five fish species (*Capoeta trutta*, *Capoeta umbla*, *Carasobarbus luteus*, *Carassius gibelio*, and *Cyprinus carpio*) from Dicle Dam Lake, Diyarbakır, in the Mesopotamian River system. *C. gibelio* exhibited the highest total lipid values, with percentages of 7.57% in winter, 8.55% in summer, and 8.30% in autumn. In *C. trutta*, Σ SFA peaked in autumn (38.64%), mainly due to C16:0 (27.08%), and Σ PUFA was the highest in winter (38.31%), with major contributors being EPA (12.21%) and C18:2 ω 6 (10.85%). *C. umbla* had peak Σ SFA in autumn (31.27%) and Σ PUFA in winter (43.06%), driven by C18:2 ω 6 (19.14%) and EPA (9.27%). Both *C. trutta* and *C. umbla* had maximum Σ MUFA in summer, mainly from C18:1 ω 9. *C. luteus* showed high Σ PUFA in autumn (48.88%) and summer (48.30%), with DHA C22:6 ω 3 reaching 16.69% in summer. *C. gibelio* had peak Σ PUFA in winter (49.89%) and summer (49.33%). *C. umbla* had the highest $\Sigma\omega$ 3/ $\Sigma\omega$ 6 ratio in spring (2.04). Seasonal variations in fatty acid composition reflect the dynamic lipid profiles of these commercially important fish, offering insights into their nutritional quality and guiding seasonal dietary recommendations.

KEY-WORDS: Dicle Dam Lake; Freshwater fish; Polyunsaturated fatty acids; Total lipid.

RESUMEN: *Cambios estacionales en los perfiles de ácidos grasos de especies de pescado ampliamente consumidas en la cuenca fluvial mesopotámica.* Este estudio investiga las variaciones estacionales en el contenido total de lípidos y la composición de ácidos grasos de cinco especies de peces (*Capoeta trutta*, *Capoeta umbla*, *Carasobarbus luteus*, *Carassius gibelio* y *Cyprinus carpio*) del embalse Dicle, Diyarbakır, en el sistema fluvial mesopotámico. *C. gibelio* mostró los valores totales de lípidos más altos, con porcentajes de 7,57 % en invierno, 8,55 % en verano y 8,30 % en otoño. En *C. trutta*, Σ SFA alcanzó su máximo en otoño (38,64 %), principalmente debido a C16:0 (27,08 %), y Σ PUFA fue más alto en invierno (38,31 %), con los principales contribuyentes siendo EPA (12,21 %) y C18:2 ω 6 (10,85 %). *C. umbla* tuvo su Σ SFA máxima en otoño (31,27 %) y Σ PUFA en invierno (43,06 %), impulsado por C18:2 ω 6 (19,14 %) y EPA (9,27 %). Tanto *C. trutta* como *C. umbla* presentaron su máximo de Σ MUFA en verano, principalmente por C18:1 ω 9. *C. luteus* mostró altos valores de Σ PUFA en otoño (48,88 %) y verano (48,30 %), con DHA C22:6 ω 3 alcanzando 16,69 % en verano. *C. gibelio* alcanzó su Σ PUFA máximo en invierno (49,89 %) y verano (49,33 %). *C. umbla* presentó la relación $\Sigma\omega$ 3/ $\Sigma\omega$ 6 más alta en primavera (2,04). Las variaciones estacionales en la composición de ácidos grasos reflejan los perfiles lipídicos dinámicos de estos peces de importancia comercial, proporcionando información sobre su calidad nutricional y orientando recomendaciones dietéticas estacionales.

PALABRAS CLAVE: Ácidos grasos poliinsaturados; Embalse Dicle; Lípido total; Peces de agua dulce.

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1. INTRODUCTION

The fatty acid composition of fish is influenced by a range of environmental and biological factors, including seasonal changes, water temperature, and diet (Özogul *et al.*, 2007). Water temperature, in particular, plays a crucial role for ectothermic species like fish, as it directly affects metabolic rates, growth, and lipid metabolism, leading to seasonal fluctuations in fatty acid profiles (Özogul *et al.*, 2007). Temperature-driven adjustments also impact feeding behavior and energy demands, contributing to shifts in lipid composition. ω 3 polyunsaturated fatty acids (PUFAs) are known to aid membrane fluidity and temperature adaptation, particularly in cold environments (Aras, 2003). Additionally, factors such as fish size, age, reproductive status, and food availability further modulate lipid content and composition, reflecting complex interactions between environmental conditions and biological requirements (Fatma and Ahmed, 2020; Guler *et al.*, 2008).

The freshwater ichthyofauna of southeastern Türkiye, particularly within the Tigris and Euphrates River systems, is highly diverse, with several endemic species of commercial and ecological significance. Species such as *Capoeta trutta* (Longspine scraper), *Capoeta umbla* (Tiger scraper), and *Carasobarbus luteus* (Yellow barbel) are native to the region, while non-native species like *Carassius gibelio* (Prussian carp) and *Cyprinus carpio* (Common carp) have been introduced through aquaculture (Cengiz *et al.*, 2010; Kaçar and Başhan, 2016). Dicle Dam Lake, located in Diyarbakır, is a vital habitat for these species, supporting both biodiversity and regional fisheries. This study examines the seasonal variations in the fatty acid profiles and total lipid contents of these species, shedding light on their nutritional value and ecological roles. The findings contribute to understanding metabolic adaptations and offer implications for the management of fisheries and for human nutrition.

2. MATERIALS AND METHODS

2.1. Sample collection and preparation

In this study, fish samples were collected from Dicle Dam Lake (38°19'53.0"N, 40°12'31.6"E), located in Diyarbakır, Turkey, during the winter (December–February), spring (March–May), summer (June–August), and autumn (September–November) seasons of 2023. A total of five fish species, including *C. trutta*, *C. umbla*, *C. luteus*, *C. gibelio*, and *C. carpio*, were targeted for this research due to their economic value and frequent consumption in the region. The collection of fish samples occurred in various seasonal sampling trips, ensuring representation of each season. The samples were obtained using a combination of fishing nets and traps, tailored to minimize stress on the fish. Each sampling event was conducted following ethical guidelines to ensure the welfare of the captured species. The collected fish were immediately placed on ice and transported to the laboratory. Upon arrival at the laboratory, morphometric measurements were taken for each specimen, including wet weight (WW) and total length (cm). Identification of the fish species was conducted by ichthyologists from the Department of Biology at Dicle University, Turkey. Specimens were selected based on uniform size criteria to ensure comparability, targeting an average of 40 individuals per species. For each season, a subset of 20 similarly sized individuals (females) per species was used for fatty acid analysis. The fish were prepared through processes such as gutting, deboning, and filleting, ensuring that the dorsal muscle portion between the dorsal fin and head was retained. This dorsal muscle sample was then stored at –20 °C prior to analysis. The seasonal characteristics, along with the range and mean ($\pm$ SD) of length and weight for female fish from Dicle Dam Lake used in the present study are presented in Table 1. All procedures involving the care and use of experimental animals adhered to the fish welfare laws and guidelines established by the Ministry of Agriculture and Rural Affairs of Türkiye. The author holds a certificate for animal use in experimental research from the Dicle University Animal Research Ethical Committee (certificate no. DÜHADEK 2017029), ensuring compliance with ethical standards for the use of animals in scientific research.

TABLE 1. The seasonal characteristics of fish species from Dicle Dam Lake. Length (cm), weight (g)

Fish	Winter		Spring		Summer		Autumn	
	Mean standard length	Mean total weight	Mean standard length	Mean total weight	Mean standard length	Mean total weight	Mean standard length	Mean total weight
<i>Capoeta trutta</i>	33±8	265±25	30±6	244±20	26±5	210±33	29±5	255±42
<i>Capoeta umbla</i>	35±7	512±43	32±7	490±23	31±9	485±55	33±6	480±35
<i>Carasobarbus luteus</i>	27±6	274±32	27±4	265±46	26±8	255±24	26±5	250±22
<i>Carassius gibelio</i>	22±5	246±20	19±7	232±40	21±8	215±32	21±7	210±23
<i>Cyprinus carpio</i>	51±7	985±52	50±12	924±62	45±13	885±66	52±6	875±52

Values are expressed as mean ± SD based on measurements from 20 different fish for each season.

TABLE 2. Seasonal variation in total lipid content (% wet weight, mean ± SD) of five fish species from Dicle Dam Lake. Different superscript letters within a row indicate significant differences among seasons ($P \leq 0.05$)

Species	Winter	Spring	Summer	Autumn
	(Mean ± SD, %)	(Mean ± SD, %)	(Mean ± SD, %)	(Mean ± SD, %)
<i>Capoeta trutta</i>	3.85 ± 0.36a	4.65 ± 0.53b	5.48 ± 0.47c	3.96 ± 0.32a
<i>Capoeta umbla</i>	7.38 ± 0.59a	3.45 ± 0.33b	4.58 ± 0.35c	4.43 ± 0.51c
<i>Carasobarbus luteus</i>	7.28 ± 0.55a	2.46 ± 0.24b	3.67 ± 0.41c	5.16 ± 0.47d
<i>Carassius gibelio</i>	7.57 ± 0.57a	2.23 ± 0.35b	8.55 ± 0.71c	8.30 ± 0.65c
<i>Cyprinus carpio</i>	2.81 ± 0.42a	2.17 ± 0.21a	2.86 ± 0.04a	5.62 ± 0.44b

2.2. Analysis of fatty acid methyl esters (FAME) and GC-FID conditions

Total lipids were extracted from the dorsal muscle tissue following the method described by Bligh and Dyer (1959), using a chloroform-methanol-water mixture (1:2:0.8, v/v/v). The lipid extracts were subsequently subjected to transesterification using acidified methanol to obtain fatty acid methyl esters (FAMES) (Ekin, 2023; Satar *et al.*, 2012).

FAMES were analyzed using a gas chromatograph (Agilent 7820A) equipped with a flame ionization detector (FID) and a DB-23 capillary column (60 m length, 0.25–0.32 mm i.d., 0.10–0.30 µm film thickness). The GC oven was thermostatically regulated with a precision of ± 0.1 °C. Prior to analysis, the column was conditioned by gradually increasing the oven temperature from ambient at a rate of 3 °C/min until a stable, linear baseline was achieved, which was then maintained for one hour.

The initial oven temperature was set to 165 °C and held for 15 minutes, then ramped to 200 °C at a rate of 5 °C/min. Injections were performed at 250 °C using a 10 µL manual injector (graduated to 0.1 µL), with a sample injection volume of 1 µL. A split ratio of 1:20 was used, optimized based on sample concentration. When trace levels of components were expected, the injected volume (initially 0.1–0.2 µL) was increased up to tenfold to enhance detectability. Nitrogen (1.2 mL/min) served as the carrier gas, with high-purity hydrogen (≥ 99.9 %) and dry air as auxiliary gases. For samples containing short-chain fatty acids (< C12), injections were made at 100 °C, with a rapid oven ramp of 4–8 °C/min to reach the target separation temperature. The program was held at isothermal conditions until complete elution of all peaks. *Trans*-isomer separation of fatty acids with carbon chains ranging from C10 to C24 was achieved using a specific cyanopropyl-silica coated capillary column of the same dimensions. Identification of FAMES was carried out by comparing their retention times with those of authentic standards (Fatty Acid Methyl Esters Standard Mixture, ME14-1KT, Sigma-Aldrich, St. Louis, MO, USA). Quantification was based on relative FID peak areas, and data acquisition and processing were conducted using Hewlett-Packard ChemStation software (model 3365). For illustration, Figure 1 presents GC-FID chromatograms from the first of three replicates in the fatty acid analysis: (A) *C. carpio* during winter (GC-FID analysis code: 6587), and (B) *C. luteus* during summer (GC-FID analysis code: 6594).

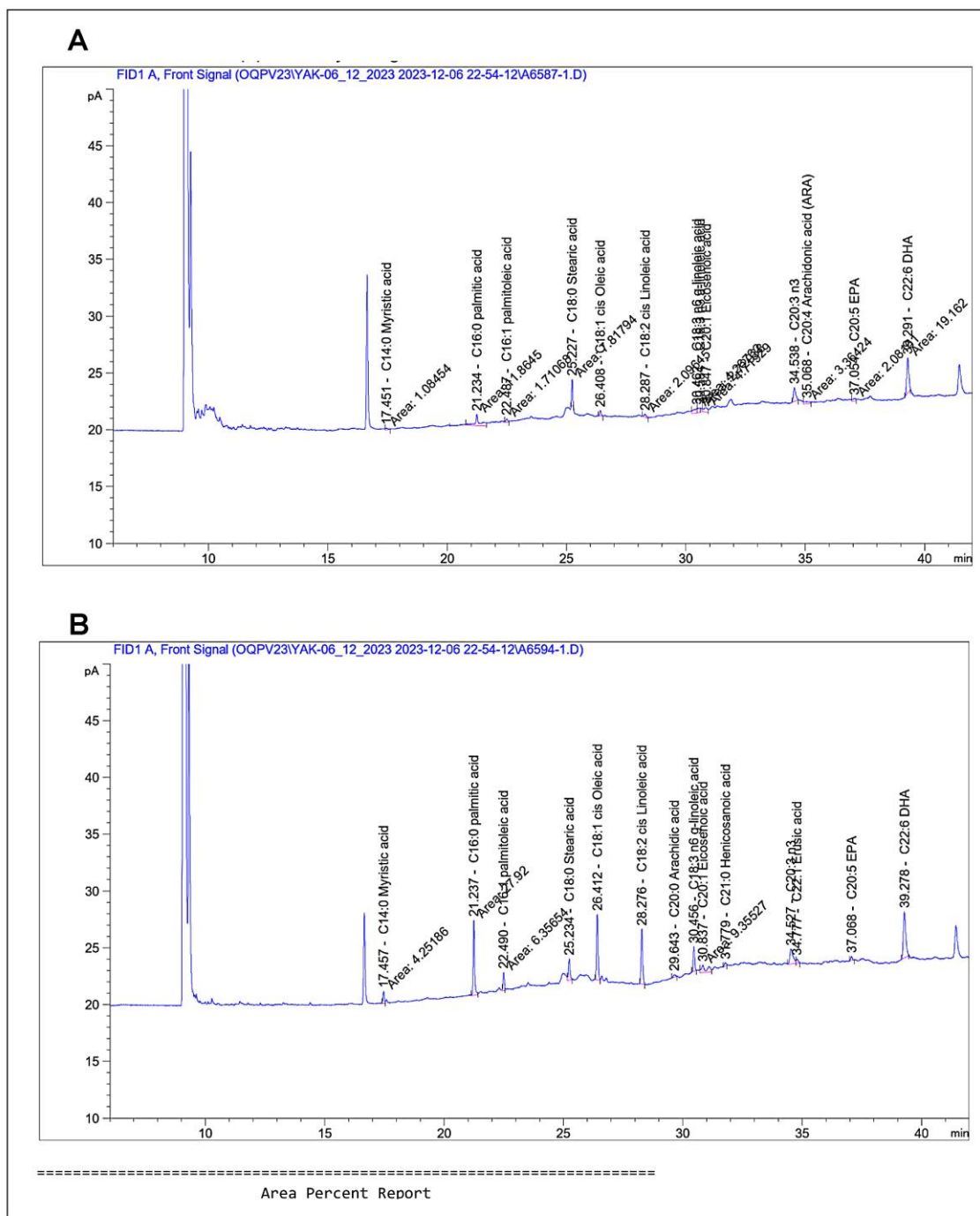


FIGURE 1. GC-FID chromatograms from the first replicate of fatty acid analysis for: (A) *Cyprinus carpio* during winter (GC-FID analysis code: 6587), and (B) *Carasobarbus luteus* during summer (GC-FID analysis code: 6594).

2.3. Statistical analyses

Data are expressed as mean values with corresponding standard deviations (mean $\pm$ SD). Statistical analyses were performed using SPSS 16.0 software. Fatty acid levels were analyzed using one-way analysis of variance (ANOVA), followed by Tukey's post-hoc test for multiple comparisons. All measurements were conducted in triplicate, and statistically significant differences were considered at a threshold of $P \leq 0.05$.

3. RESULTS AND DISCUSSION

In this study, a total of 37 fatty acids were analyzed. However, C4:0, C6:0, C8:0, C11:0, C13:0, C18:1 ω 9-*trans*, C18:2 ω 6-*trans*, and C22:2 ω 6 were not detected in all fish species across four seasons (Tables 3–7).

3.1. Seasonal variation in total lipid content in the fish

Seasonal variations in water temperature significantly influence lipid metabolism in fish. Colder winter months typically result in increased lipid accumulation due to lower metabolic rates and energy storage, whereas warmer summer months lead to energy expenditure and reduced lipid reserves (Balıkçı, 2021; Fernandes and McMeans, 2019). Seasonal nutrient availability also impacts lipid storage and utilization (Ekin, 2023; Tian *et al.*, 2016).

In the current study, total lipid content varied significantly across species and seasons ($P \leq 0.05$). *C. trutta* exhibited its highest lipid content in summer (5.48%) and lowest in winter (3.85%). *C. umbla* reached its peak in winter (7.38%) and minimum in spring (3.45%), with intermediate levels in summer (4.58%) and autumn (4.43%). *C. luteus* showed the highest lipid content in winter (7.28%) and lower levels in spring (2.46%), summer (3.67%), and autumn (5.16%). *C. gibelio* recorded maximum lipid content in summer (8.55%) and autumn (8.30%), with the lowest value in spring (2.23%). Conversely, *C. carpio* peaked in autumn (5.62%) and had its lowest lipid level in spring (2.17%). These findings are consistent with previous studies reporting seasonal lipid variations in similar taxa. For example, *C. carpio* showed lipid contents of 0.88% in Beyşehir Lake (Özogul *et al.*, 2007), 3.33% in winter (Öksüz *et al.*, 2019), and 10.07% in market fish in Serbia during winter (Ljubojevic *et al.*, 2013). Semi-intensive cultured *C. carpio* in the Czech Republic had 6.48% lipid content (Linhartova *et al.*, 2018), while seasonal fluctuations in Beyşehir Lake ranged from 1.09% in summer to 4.45% in winter (Guler *et al.*, 2008). Compared to these reports, the current study showed generally higher lipid values in *C. umbla* and *C. luteus* during winter, while *C. trutta* and *C. carpio* exhibited similar seasonal patterns but with some variability likely due to regional differences in food availability and environmental conditions.

Differences in lipid content between studies can be attributed to factors such as seasonal food availability, species-specific feeding habits, metabolic demands, and environmental conditions, including water temperature, salinity, and oxygen levels (Guler *et al.*, 2008; Kaçar *et al.*, 2018). The omnivorous feeding behavior of *C. trutta*, *C. umbla*, *C. luteus*, *C. gibelio*, and *C. carpio*, combined with seasonal prey abundance and food diversity, likely drives these variations. The deep, stratified nature of Dicle Dam Lake further influences lipid metabolism and storage, reflecting the species' adaptive responses to environmental conditions (Emre *et al.*, 2020; Memon *et al.*, 2010; Satar *et al.*, 2012; Tian *et al.*, 2016).

Fish are generally categorized by their total lipid content as lean fish (lipid content less than 2%), low-fat fish (lipid content 2–4%), medium-fat fish (lipid content 4–8%), and high-fat fish (lipid content above 8%) (Ekin, 2023; Kaçar *et al.*, 2018). The seasonal variations in total lipid content in the fish species in this study show differences that align with established lipid categories for fish. *C. trutta* had its highest lipid content in summer (5.48%), placing it in the medium-fat category, while it remained low-fat in other seasons. *C. umbla* accumulated more lipid in winter (7.38%), positioning it as medium-fat during colder months and low-fat in spring (3.45%). *C. luteus* showed a similar trend, with medium-fat levels in winter (7.28%) and lower lipid content in spring (2.46%). *C. gibelio* reached high-fat status in summer (8.55%) and autumn (8.30%), reflecting its lipid storage during warmer months, but dropped to low-fat in spring (2.23%). *C. carpio* was medium-fat in autumn (5.62%), though it remained low-fat in winter and spring.

TABLE 3. Seasonal fatty acid composition of the total lipids from the dorsal muscle of *Capoeta trutta* (% of total fatty acids)

Fatty Acids (%)	Winter	Spring	Summer	Autumn
C4:0	nd	nd	nd	nd
C6:0	nd	nd	nd	nd
C8:0	nd	nd	nd	nd
C10:0	nd	nd	nd	nd
C11:0	nd	nd	nd	nd
C12:0	nd	nd	nd	nd
C13:0	nd	nd	nd	nd
C14:0	3.59±0.02a	3.98±0.06a	3.50±0.01a	3.16±0.06a
C15:0	nd	nd	0.65±0.01a	1.07±0.01b
C16:0	18.60±0.38a	18.61±0.42a	19.99±0.14a	27.08±0.05b
C17:0	nd	nd	0.42±0.00a	0.46±0.04a
C18:0	10.37±0.18a	5.67±0.14b	3.06±0.03c	6.22±0.01b
C20:0	nd	5.42±0.83a	0.83±0.01b	nd
C21:0	nd	1.89±0.06a	1.80±0.12a	0.52±0.03b
C22:0	nd	nd	0.25±0.01a	0.13±0.19a
C23:0	nd	nd	nd	nd
C24:0	1.15±0.10	nd	nd	nd
C14:1ω5	0.10±0.00a	nd	0.29±0.01a	0.47±0.04a
C15:1ω5	nd	nd	0.07±0.00	nd
C16:1ω7	2.62±0.06a	18.36±0.33b	19.70±0.10b	12.94±0.05c
C17:1ω7	nd	4.27±0.14a	4.03±0.01a	1.24±0.02b
C18:1ω9- <i>trans</i>	nd	nd	nd	nd
C18:1ω9	23.73±0.37a	9.51±0.25b	16.63±0.05c	23.12±0.02a
C20:1ω9	1.79±0.28a	1.42±0.36a	0.36±0.01b	1.30±0.02a
C22:1ω9	0.20±0.14	nd	nd	nd
C24:1ω9	nd	nd	nd	nd
C18:2ω6- <i>trans</i>	nd	nd	nd	nd
C18:2ω6	10.85±0.15a	2.99±0.06b	4.07±0.01c	8.61±0.01d
C18:3ω6	5.34±0.34a	8.04±0.46b	4.71±0.03a	2.48±0.14c
C18:3ω3	5.22±0.13a	2.25±0.02b	5.93±0.02a	2.78±0.04b
C20:2ω6	0.91±0.29a	nd	0.22±0.00a	0.37±0.02a
C20:3ω6	1.06±0.00a	nd	0.25±0.01b	0.48±0.02b
C20:3ω3	nd	nd	nd	nd
C20:4ω6	1.16±0.10a	1.19±0.03a	1.00±0.08a	1.44±0.02a
C20:5ω3	12.21±0.10a	13.13±0.56a	9.76±0.02b	2.72±0.01a
C22:2ω6	nd	nd	nd	nd
C22:6ω3	1.56±0.08a	3.18±0.16b	2.45±0.01c	3.47±0.06b
ΣSFA	33.71±0.33a	35.57±0.73b	30.50±0.08c	38.64±0.19d
ΣMUFA	28.44±0.12a	33.56±0.87b	41.08±0.04c	39.07±0.20c
ΣPUFA	38.31±0.48a	30.78±0.98b	28.39±0.06b	22.35±0.15c
Σω6	19.32±0.08a	12.22±0.60b	10.25±0.03c	13.38±0.08b
Σω3	18.99±0.40a	18.56±0.38a	18.14±0.03a	8.97±0.07b
Σω3/Σω6	0.98	1.52	1.77	0.67

Values are expressed as means ± SD for three replicates and are reported as percentages of total fatty acid methyl esters. The mean values of different characters within the same row were compared using one-way ANOVA followed by Tukey's post-hoc test. Different letters indicate significant differences ($P \leq 0.05$). Results are expressed as % fatty acids of total lipids. nd – not detected.

3.2. Seasonal variation in fatty acid composition of *Capoeta trutta*

The seasonal variation in the fatty acid profile of *Capoeta trutta* is summarized in Table 3. Satar *et al.* (2012) examined the seasonal variation in fatty acid composition in the dorsal muscle of *C. trutta* from the Tigris River, Diyarbakır, Turkey, revealing peak levels of C16:1 ω 7, C18:2 ω 6, C18:3 ω 3, and C20:5 ω 3, linked to favorable feeding conditions. The current study's results show both similarities and discrepancies. In this study, Σ SFA (saturated fatty acid) was dominant in autumn (38.64%), with C16:0 increasing from 18.60% in winter to 27.08% in autumn. C18:0 levels were notably lower in summer (3.06%) compared to other seasons (winter: 10.37%, spring: 5.67%, autumn: 6.22%). C18:1 ω 9 exhibited seasonal fluctuations, peaking in winter (23.73%) and autumn (23.12%) while reaching its lowest in spring (9.51%). Satar *et al.* (2012) reported peak Σ SFA and Σ MUFA (monounsaturated fatty acids) levels in summer, predominantly due to C16:0 and C18:1 ω 9, but in this study, Σ SFA peaked in autumn, driven by C16:0, suggesting that environmental factors may impact seasonal profiles differently. Both studies observed peak Σ MUFA in summer, attributed to C16:1 ω 7 (19.70%) and C18:1 ω 9 (16.63%) in this study, indicating some consistency. While Satar *et al.* (2012) found peak Σ PUFA (polyunsaturated fatty acids) in autumn (57.78%), our results showed that Σ SPUFA peaked in winter (38.31%), with C18:2 ω 6 and C20:5 ω 3 as the primary contributors. Additionally, Ekin (2023) reported that total lipids in *C. trutta* were largely composed of Σ SFA (52.06%), with C16:0 being the most abundant in summer. Σ MUFA accounted for 46.29% of total fatty acids, with C16:1 ω 7 as the most prominent, and Σ PUFA was found in low levels (1.68%) Ekin (2023). The seasonal variations in fatty acids observed in this study may be attributed to the dietary habits of *C. trutta*, particularly zooplankton and phytoplankton abundance, which peak in spring and decline in winter (Balıkcı, 2021). The low ω 3 PUFA content (8.97%) in autumn could be due to decreased consumption of zooplankton and phytoplankton, which are rich in PUFAs like C20:5 ω 3 and C22:6 ω 3 (Kaçar and Başhan, 2016; Steffens, 1997). Furthermore, the lower accumulation of ω 3 PUFAs in autumn may be linked to the reduced availability of C18:3 ω 3, a precursor for synthesizing higher-chain PUFAs such as C22:6 ω 3. The prevalence of ω 6 fatty acids in freshwater fish diets, rich in algae, insect larvae, and crustaceans, may explain the elevated levels of C18:2 ω 6 and C18:3 ω 6 (Kaçar and Başhan, 2016; Steffens, 1997).

3.3. Seasonal variation in the fatty acid composition of *Capoeta umbla*

The seasonal variation in fatty acids in *C. umbla* is presented in Table 4. In the current study, significant seasonal variations were observed in the fatty acid composition of *C. umbla*, with marked fluctuations in Σ SFA, Σ MUFA, and Σ PUFA levels throughout the year. The highest Σ SFA content was recorded in autumn at 31.27%, mainly driven by an increase in C16:0, which rose significantly from 15.96% in winter to 23.28% in autumn ($p \leq 0.05$). This autumnal elevation in C16:0 could be attributed to seasonal dietary shifts or metabolic adaptations, aligning with Kaçar *et al.* (2023), who reported elevated SFA levels in January preceding the reproductive season. Aras (2009) similarly reported seasonal increases between January and February in Σ SFA (42.05–31.71%) for *C. capoeta umbla* across various habitats, emphasizing how environmental conditions in rivers and reservoirs contribute to Σ SFA variability. In contrast, Ekin, (2023) reported a higher Σ SFA level of 46.79% in summer, with C16:0 as the predominant fatty acid, while a study by Çelik *et al.* (2005) recorded a peak Σ SFA of 42.05% during summer in the Tuzla River population. These findings suggest that environmental factors such as seasonal temperature changes and food availability can significantly influence lipid metabolism and fatty acid profiles in freshwater fish, with variations across habitats shaping the observed fatty acid levels (Çelik *et al.*, 2005).

TABLE 4. Seasonal fatty acid composition of the total lipids from the dorsal muscle of *Capoeta umbla* (% of total fatty acids)

Fatty Acids (%)	Winter	Spring	Summer	Autumn
C4:0	nd	nd	nd	nd
C6:0	nd	nd	nd	nd
C8:0	nd	nd	nd	nd
C10:0	nd	nd	nd	nd
C11:0	nd	nd	nd	nd
C12:0	nd	nd	nd	nd
C13:0	nd	nd	nd	nd
C14:0	2.93±0.01a	6.57±0.40b	1.30±0.01c	1.56±0.11c
C15:0	nd	0.27±0.01a	nd	0.92±0.03a
C16:0	15.96±0.02a	18.42±0.26b	16.33±0.12a	23.28±0.32c
C17:0	0.32±0.01a	0.71±0.01a	nd	nd
C18:0	2.22±0.01a	3.91±0.04b	2.66±0.12a	3.97±0.13b
C20:0	1.47±0.00a	0.67±0.02b	nd	1.07±0.02a
C21:0	0.58±0.07a	0.17±0.01a	1.16±0.02b	0.47±0.01a
C22:0	0.52±0.00a	0.27±0.02a	nd	nd
C23:0	nd	nd	nd	nd
C24:0	nd	nd	nd	nd
C14:1ω5	0.33±0.01a	0.20±0.01a	0.52±0.00a	0.46±0.02a
C15:1ω5	0.05±0.01a	nd	0.05±0.03a	nd
C16:1ω7	9.23±0.01a	14.66±0.28b	14.72±0.01b	19.00±0.52c
C17:1ω7	0.96±0.02a	3.20±0.02b	1.86±0.13c	2.40±0.14bc
C18:1ω9- <i>trans</i>	nd	nd	nd	nd
C18:1ω9	21.95±0.03a	12.02±0.06b	25.65±0.05c	16.06±0.20d
C20:1ω9	0.05±0.01a	3.38±0.08b	2.55±0.00c	1.74±0.01d
C22:1ω9	nd	nd	nd	nd
C24:1ω9	nd	1.00±0.00a	nd	nd
C18:2ω6- <i>trans</i>	nd	nd	nd	nd
C18:2ω6	19.14±0.01a	2.87±0.04b	10.75±0.02c	5.05±0.07d
C18:3ω6	6.06±0.01a	7.40±0.03b	6.23±0.01a	6.50±0.01a
C18:3ω3	4.82±0.02a	8.37±0.13b	5.95±0.01c	3.80±0.08d
C20:2ω6	0.45±0.01	nd	nd	nd
C20:3ω6	nd	0.27±0.02	nd	nd
C20:3ω3	nd	1.62±0.06	nd	nd
C20:4ω6	1.24±0.04a	0.99±0.06a	2.23±0.09b	0.98±0.02a
C20:5ω3	9.27±0.01a	10.74±0.31a	6.01±0.01b	9.4±0.13a
C22:2ω6	nd	nd	nd	nd
C22:6ω3	2.08±0.09a	2.80±0.21a	2.29±0.01a	3.18±0.12a
ΣSFA	24.00±0.05a	30.99±0.18b	21.45±0.01c	31.27±0.21b
ΣMUFA	32.57±0.07a	34.36±0.28b	45.35±0.12c	39.66±0.43d
ΣPUFA	43.06±0.13a	35.06±0.26b	33.46±0.13b	28.91±0.38c
Σω6	26.89±0.10a	11.53±0.16b	19.21±0.10c	12.53±0.07b
Σω3	16.17±0.03a	23.53±0.10b	14.25±0.03c	16.38±0.31a
Σω3/Σω6	0.60	2.04	0.74	1.31

Values are expressed as means ± SD for three replicates and are reported as percentages of total fatty acid methyl esters. The mean values of different characters within the same row were compared using one-way ANOVA followed by Tukey's post-hoc test. Different letters indicate significant differences ($P \leq 0.05$). Results are expressed as % fatty acids of total lipids. nd – not detected.

In the current study, in terms of Σ MUFA, the summer season exhibited the highest concentration at 45.35%, largely due to increased levels of C18:1 ω 9 and C16:1 ω 7 ($p \leq 0.05$). Kaçar *et al.* (2023), noted increased MUFA concentrations in January (38.96%) prior to reproduction, with seasonal declines suggesting a metabolic shift in lipid utilization associated with reproductive cycles. The significant increase in C18:1 ω 9 content from spring (12.02%) to summer highlights the influence of seasonal dietary shifts, likely resulting from increased intake of zooplankton and phytoplankton during warmer months, known to be rich in MUFAs (Balıkçı, 2021). Similar trends were reported in fish from Tercan Dam Lake, where Σ MUFA levels peaked in summer (Çelik *et al.*, 2005).

Interestingly, in the current study, the highest levels of Σ PUFA were detected in winter (43.06%), with C18:2 ω 6 and C20:5 ω 3 making significant contributions. However, contrary to initial assumptions, the elevated levels of C18:2 ω 6 in winter (19.14%) may not be directly linked to zooplankton and phytoplankton reservoirs, as zooplankton populations typically peak in spring and summer rather than winter. This discrepancy suggests that other dietary sources or metabolic adaptations during colder months could account for the higher Σ PUFA levels in winter (Memon *et al.*, 2010). The seasonal fluctuations in C18:3 ω 3 and C18:3 ω 6 also align with observations by Kaçar *et al.* (2023), where triacylglycerol and phospholipid compositions showed varying levels of essential fatty acids, depending on reproductive and metabolic states. Additionally, the significant seasonal decline of C18:2 ω 6 from 19.14% in winter to 2.87% in spring ($p \leq 0.05$) for the present study suggests that environmental or dietary factors strongly influence PUFA composition. Kaçar *et al.* (2023) similarly observed significant seasonal Σ PUFA variation, with a notable shift in muscle tissue PUFAs, indicating that *C. umbla* may undergo adaptive changes to optimize PUFA profiles seasonally. In the current study, it was observed that C18:3 ω 3 peaked in spring (8.37%), indicating an increase in ω 3-rich dietary intake or endogenous biosynthesis during this period. Another noteworthy finding is the favourable ω 3/ ω 6 ratio, particularly in spring (2.04), indicating a diet rich in ω 3 PUFAs. This is significant for the ecological dynamics of *C. umbla* and its potential nutritional benefits, as diets with higher $\Sigma\omega$ 3/ $\Sigma\omega$ 6 ratios are associated with reduced risks of chronic diseases, including cardiovascular conditions and inflammation (Simopoulos, 2002). In comparison to the Tuzla River population, which exhibiting higher Σ SFA levels in summer, the $\Sigma\omega$ 3/ $\Sigma\omega$ 6 ratio remained relatively stable throughout the year (Aras, 2003). This stability further emphasizes the influence of local environmental conditions on the fatty acid profiles of *C. umbla*. The composition of fatty acids and the ω 3/ ω 6 ratio are influenced by the diet consumed by fish (Steffens, 1997). Kaçar *et al.* (2023) reported similarly high $\Sigma\omega$ 3/ $\Sigma\omega$ 6 ratios in muscle tissue during the summer reproductive season, highlighting that such fluctuations may correlate with reproductive requirements or available prey which is rich in ω 3 PUFAs. Historical research suggests that early human diets maintained an $\Sigma\omega$ 6/ $\Sigma\omega$ 3 ratio close to 1:1, while modern western diets exhibit a markedly higher ratio of 15–17:1 (Çelebi *et al.*, 2017). In contrast, elevated levels of ω 3 PUFA have been associated with protective health effects (Simopoulos, 2002). An optimal $\Sigma\omega$ 6/ $\Sigma\omega$ 3 ratio for health benefits is proposed to be between 1:1 and 2:1 (Simopoulos, 2002).

3.4. Seasonal variation in fatty acid composition of *Carasobarbus luteus*

Table 5 presents the seasonal variation in the fatty acid composition of *C. luteus*. In this study, the Σ SFA levels in *C. luteus* were the highest during winter (35.63%), predominantly driven by the abundance of C16:0, which peaked at 24.78% ($p \leq 0.05$). These results align with previous findings, wherein C16:0 was also the predominant SFA in *C. luteus*, albeit at lower concentrations; for example, Cengiz *et al.*, (2010) reported that C16:0 constituted 12.28% of the total fatty acids in winter, a value significantly lower than that observed in the present study. This discrepancy may stem from geographic and environmental differences, alongside seasonal sampling variations.

TABLE 5. Seasonal fatty acid composition of the total lipids from the dorsal muscle of *Carasobarbus luteus* (% of total fatty acids)

Fatty Acids (%)	Winter	Spring	Summer	Autumn
C4:0	nd	nd	nd	nd
C6:0	nd	nd	nd	nd
C8:0	nd	nd	nd	nd
C10:0	nd	nd	nd	nd
C11:0	nd	nd	nd	nd
C12:0	nd	nd	nd	nd
C13:0	nd	nd	nd	nd
C14:0	2.56±0.26a	1.36±0.47b	2.59±0.11a	0.90±0.00b
C15:0	0.77±0.00a	0.29±0.02a	nd	0.39±0.01a
C16:0	24.78±0.30a	19.16±0.07b	16.74±0.72c	13.61±0.02d
C17:0	0.86±0.00a	0.25±0.02a	nd	0.21±0.00a
C18:0	5.15±0.04a	4.98±0.01a	6.55±0.02b	2.92±0.01c
C20:0	nd	0.42±0.01a	1.01±0.04a	0.70±0.01a
C21:0	1.37±0.01a	0.80±0.01a	2.79±0.42b	0.87±0.02a
C22:0	0.14±0.01a	0.54±0.01a	nd	0.67±0.00a
C23:0	nd	nd	nd	nd
C24:0	nd	nd	nd	nd
C14:1ω5	nd	0.23±0.03a	nd	0.19±0.01a
C15:1ω5	nd	nd	nd	nd
C16:1ω7	10.85±0.10a	4.24±0.04b	3.87±0.05b	2.99±0.01c
C17:1ω7	1.07±0.01a	0.35±0.01b	nd	0.34±0.01b
C18:1ω9- <i>trans</i>	nd	nd	nd	nd
C18:1ω9	20.95±0.09a	27.27±0.07b	17.38±0.33c	25.06±0.04d
C20:1ω9	1.35±0.01a	nd	0.49±0.69b	1.96±0.01a
C22:1ω9	nd	nd	nd	nd
C24:1ω9	nd	nd	nd	nd
C18:2ω6- <i>trans</i>	nd	nd	nd	nd
C18:2ω6	6.26±0.06a	19.60±0.03b	15.23±0.05c	25.02±0.01d
C18:3ω6	5.34±0.02a	15.56±0.04b	6.71±0.13c	4.20±0.00a
C18:3ω3	4.49±0.03a	1.79±0.04b	1.61±0.07b	6.72±0.01c
C20:2ω6	0.51±0.01a	0.33±0.02a	nd	0.37±0.02a
C20:3ω6	0.44±0.01a	0.54±0.01a	nd	nd
C20:3ω3	3.66±0.01a	0.61±0.01b	5.82±0.16c	0.66±0.01b
C20:4ω6	1.67±0.02a	0.38±0.01b	1.03±0.07a	1.49±0.12a
C20:5ω3	6.26±0.04a	1.12±0.01b	1.21±0.05b	9.14±0.01c
C22:2ω6	nd	nd	nd	nd
C22:6ω3	2.31±0.01a	0.19±0.04b	16.69±0.02c	1.28±0.01d
ΣSFA	35.63±0.29a	27.80±0.47b	29.68±0.81b	20.27±0.05c
ΣMUFA	34.22±0.19a	32.09±0.06b	21.74±0.31c	30.54±0.05d
ΣPUFA	30.94±0.15a	40.12±0.08b	48.30±0.10c	48.88±0.10c
Σω6	14.22±0.06a	36.41±0.07b	22.97±0.04c	31.08±0.08d
Σω3	16.72±0.09a	3.71±0.01b	25.33±0.05c	17.80±0.02a
Σω3/Σω6	1.18	0.11	1.10	0.57

Values are expressed as means ± SD for three replicates and are reported percentages of total fatty acid methyl esters. The mean values of different characters within the same row were compared using one-way ANOVA followed by Tukey's post-hoc test. Different letters indicate significant differences ($P \leq 0.05$). Results are expressed as % fatty acids of total lipids. nd – not detected.

The Σ MUFA content in the current study reached its zenith in winter (34.22%) and spring (32.09%) influenced primarily by C18:1 ω 9 and C16:1 ω 7, which comprised a substantial portion of the total lipids. In particular, C18:1 ω 9 exhibited high levels in spring (27.27%) and showed significant variability throughout the year. In addition, during the winter period, this fatty acid level was 20.95%, which was in agreement with Cengiz *et al.* (2010) who recorded winter levels at 19.64%. In comparison, the summer MUFA content in Ekin (2023) was higher (39.44%), with C18:1 ω 9 reaching 28.81%, consistent with the peak observed in spring (27.27%) in this study, likely resulting from increased intake of zooplankton and phytoplankton during warmer months, which are known to be rich in MUFAs (Balıkçı, 2021).

The most pronounced seasonal variation was evident in Σ PUFA levels, which peaked in autumn (48.88%) due to significant contributions mainly from C18:2 ω 6, and in summer (48.30%) due to the contribution from DHA, C22:6 ω 3. The high concentration of DHA, C22:6 ω 3, particularly in summer (16.69%) (confirmed through GC–FID analysis, code: 6594, Figure 1B), highlights the nutritional richness of *C. luteus* during this period. DHA, C22:6 ω 3 is a critical ω 3 fatty acid recognized for its role in brain function, cardiovascular health, and anti-inflammatory properties. Therefore, consuming this fish will provide significant health benefits. In contrast, Cengiz *et al.* (2010) also reported higher levels of DHA, C22:6 ω 3 during winter (11.97%), while DHA–C22:6 ω 3 was not specifically mentioned in Ekin (2023), further underscoring its prominence in the current study's seasonal analysis. C18:2 ω 6 peaked at 25.02% in autumn, markedly exceeding the 8.70% reported by Cengiz *et al.* (2010) for winter and the 1.58% from Ekin (2023), demonstrating the significant impact of seasonal variation on the PUFA profile. From a health perspective, the increased availability of DHA, C22:6 ω 3 and C18:2 ω 6 during the autumn and winter seasons positions *C. luteus* as a valuable dietary source of essential fatty acids, contributing to improved heart health and overall well-being when consumed regularly. Additionally, EPA, C20:5 ω 3 exhibited seasonal variability, peaking in autumn (9.14%) and winter (6.26%), with values comparable to the 6.87% recorded by Cengiz *et al.* (2010). Notably, EPA was undetected in the summer analysis conducted by Ekin (2023). The seasonal $\Sigma\omega$ 3/ $\Sigma\omega$ 6 ratio revealed considerable variation, with the highest ratio observed in winter (1.18) and the lowest in spring (0.11). This finding is consistent with Cengiz *et al.* (2010), who reported a relatively balanced ratio of 1.11 during winter. However, the ratio observed in the current summer data (1.10) was markedly higher than the 0.16 ratio reported by Ekin (2023), indicating a more balanced presence of ω 3 fatty acids during summer in the present study. Freshwater fish, like many other species, can convert ALA, C18:3 ω 3, an omega-3 fatty acid, into longer-chain polyunsaturated fatty acids (ω 3 PUFAs) through chain elongation and desaturation. This metabolic pathway enables fish to synthesize essential fatty acids, such as EPA, C20:5 ω 3 and DHA, C22:6 ω 3, from ALA, which is typically found in other sources (Memon *et al.*, 2010). Additionally, the level of ω 3 fatty acids, especially C20:5 ω 3 and C22:6 ω 3 are high in fish living in cold climates and vice versa (Çelik *et al.*, 2005). Another possible explanation for the low accumulation of ω 3 and ω 6 PUFAs in fish might be the limited availability of C18:3 ω 3 and C18:2 ω 6, which are essential for the biosynthesis of C22:6 ω 3, C20:4 ω 6, and C20:5 ω 3 (Ekin, 2023). The higher proportion of ω 6 fatty acids often observed in freshwater fish could be due to dietary sources such as freshwater algae, insect larvae, and crustaceans which contain high levels of C18:2 ω 6 and C18: (Kaçar and Başhan, 2016; Steffens, 1997). However, due to the lack of D12 and D15 desaturase enzymes, fish are unable to convert C18:1 ω 9 into C18:2 ω 6 and further into C18:3 ω 3 (Tian *et al.*, 2016). Consequently, fish have an absolute requirement for ω 3 and ω 6 essential fatty acids, which must be obtained through their diet (Tian *et al.*, 2016).

3.5. Seasonal variation in the fatty acid composition of *Carassius gibelio*

Table 6 presents the seasonal variation in the fatty acid composition of *C. gibelio*. The current study reveals significant seasonal variation in the fatty acid composition of *C. gibelio*, particularly in the proportions of SFAs, MUFAs, and PUFAs. Notably, Σ SFA levels peaked in spring (30.21%) and autumn (27.77%), with palmitic acid (C16:0) being the predominant SFA, reaching maximum levels of 19.35% in spring and 16.74% in autumn. In contrast, Cakmak *et al.* (2012) reported lower Σ SFA levels (24.56%) in spring, where C16:0 accounted for 15.27%. Furthermore, Ekin (2023) noted a significantly higher Σ SFA content in summer (46.72%), with C16:0 contributing 30.2%, indicating pronounced seasonal and geographical variability.

Σ MUFA levels were the highest in spring (42.34%), primarily influenced by C18:1 ω 9, which ranged from 20.62% in autumn to 27.90% in spring. These findings contrast with those of Cakmak *et al.* (2012), who reported lower Σ MUFA levels in spring (31.45%), with C18:1 ω 9 comprising 17.40%. Ekin (2023) further documented

elevated Σ MUFA levels (46.92%) in summer, with C18:1 ω 9 reaching 29.13%. These disparities underscore the significant influence of seasonal and environmental factors on the MUFA concentrations in *C. gibelio* ($p \leq 0.05$).

TABLE 6. Seasonal fatty acid composition of the total lipids from the dorsal muscle of *Carassius gibelio* (% of total fatty acids)

Fatty Acids (%)	Winter	Spring	Summer	Autumn
C4:0	nd	nd	nd	nd
C6:0	nd	nd	nd	nd
C8:0	nd	nd	nd	nd
C10:0	nd	nd	nd	nd
C11:0	nd	nd	nd	nd
C12:0	nd	nd	nd	0.50 $\pm$ 0.01
C13:0	nd	nd	nd	nd
C14:0	1.86 $\pm$ 0.01a	2.80 $\pm$ 0.01b	1.75 $\pm$ 0.12a	2.87 $\pm$ 0.01b
C15:0	nd	0.48 $\pm$ 0.02a	0.44 $\pm$ 0.01a	0.81 $\pm$ 0.01a
C16:0	14.07 $\pm$ 0.02a	19.35 $\pm$ 0.09b	10.49 $\pm$ 0.02c	16.74 $\pm$ 0.03d
C17:0	nd	nd	0.53 $\pm$ 0.01	nd
C18:0	4.70 $\pm$ 0.01a	6.33 $\pm$ 0.03b	3.31 $\pm$ 0.06c	4.97 $\pm$ 0.02a
C20:0	0.83 $\pm$ 0.10a	nd	nd	0.46 $\pm$ 0.02a
C21:0	0.05 $\pm$ 0.07a	0.67 $\pm$ 0.02b	0.79 $\pm$ 0.01b	0.37 $\pm$ 0.02b
C22:0	nd	0.58 $\pm$ 0.02a	0.91 $\pm$ 0.01a	1.05 $\pm$ 0.01a
C23:0	0.11 $\pm$ 0.08	nd	nd	nd
C24:0	nd	nd	nd	nd
C14:1 ω 5	0.38 $\pm$ 0.02a	nd	nd	0.24 $\pm$ 0.04a
C15:1 ω 5	nd	nd	nd	nd
C16:1 ω 7	4.63 $\pm$ 0.02a	11.44 $\pm$ 0.06b	4.01 $\pm$ 0.01a	6.21 $\pm$ 0.01c
C17:1 ω 7	0.45 $\pm$ 0.01a	1.30 $\pm$ 0.02b	0.57 $\pm$ 0.01a	1.26 $\pm$ 0.02b
C18:1 ω 9- <i>trans</i>	nd	nd	nd	nd
C18:1 ω 9	21.77 $\pm$ 0.01a	27.90 $\pm$ 0.08b	26.44 $\pm$ 0.06b	20.62 $\pm$ 0.05a
C20:1 ω 9	1.36 $\pm$ 0.01a	1.70 $\pm$ 0.05a	1.30 $\pm$ 0.01a	1.62 $\pm$ 0.02a
C22:1 ω 9	nd	nd	nd	nd
C24:1 ω 9	nd	nd	nd	nd
C18:2 ω 6- <i>trans</i>	nd	nd	nd	nd
C18:2 ω 6	18.94 $\pm$ 0.16a	11.35 $\pm$ 0.06b	25.83 $\pm$ 0.10c	19.11 $\pm$ 0.04a
C18:3 ω 6	6.84 $\pm$ 0.01a	3.77 $\pm$ 0.01b	7.97 $\pm$ 0.01c	6.53 $\pm$ 0.01a
C18:3 ω 3	7.81 $\pm$ 0.01a	1.61 $\pm$ 0.03b	5.86 $\pm$ 0.01c	4.90 $\pm$ 0.02c
C20:2 ω 6	1.07 $\pm$ 0.00a	0.80 $\pm$ 0.01a	1.22 $\pm$ 0.04a	1.68 $\pm$ 0.07a
C20:3 ω 6	0.85 $\pm$ 0.04a	0.57 $\pm$ 0.02a	0.30 $\pm$ 0.04a	1.05 $\pm$ 0.01a
C20:3 ω 3	nd	2.13 $\pm$ 0.06a	nd	1.74 $\pm$ 0.02a
C20:4 ω 6	1.69 $\pm$ 0.01a	nd	1.75 $\pm$ 0.01a	nd
C20:5 ω 3	10.26 $\pm$ 0.01a	5.89 $\pm$ 0.05b	4.27 $\pm$ 0.00c	5.03 $\pm$ 0.01b
C22:2 ω 6	nd	nd	nd	nd
C22:6 ω 3	2.43 $\pm$ 0.11a	1.21 $\pm$ 0.16b	2.13 $\pm$ 0.01a	2.61 $\pm$ 0.07a
ΣSFA	21.62 $\pm$ 0.10a	30.21 $\pm$ 0.13b	18.22 $\pm$ 0.04c	27.77 $\pm$ 0.02d
ΣMUFA	28.59 $\pm$ 0.03a	42.34 $\pm$ 0.17b	32.32 $\pm$ 0.05c	29.95 $\pm$ 0.01a
ΣPUFA	49.89 $\pm$ 0.20a	27.33 $\pm$ 0.08b	49.33 $\pm$ 0.06a	42.65 $\pm$ 0.04c
$\Sigma\omega$ 6	29.39 $\pm$ 0.12a	16.49 $\pm$ 0.02b	37.07 $\pm$ 0.04c	28.37 $\pm$ 0.03a
$\Sigma\omega$ 3	20.50 $\pm$ 0.08a	10.84 $\pm$ 0.06b	12.26 $\pm$ 0.02c	14.28 $\pm$ 0.01d
$\Sigma\omega$ 3/ $\Sigma\omega$ 6	0.70	0.66	0.33	0.50

Values are expressed as means $\pm$ SD for three replicates and are reported as percentages of total fatty acid methyl esters. The mean values of different characters within the same row were compared using one-way ANOVA followed by Tukey's post-hoc test. Different letters indicate significant differences ($P \leq 0.05$). Results are expressed as % fatty acids of total lipids. nd – not detected.

For PUFAs, the current study identified Σ PUFA levels at their highest in winter (49.89%) and summer (49.33%), primarily due to substantial contributions from C18:2 ω 6 and C20:5 ω 3. In comparison, Cakmak *et al.* (2012) found lower Σ PUFA levels in spring (43.99%), with a higher contribution from C22:6 ω 3 (11.03%) than observed in the present study. Conversely, Ekin (2023) reported much lower Σ PUFA levels (6.38%) in summer, where C18:2 ω 6 contributed 3.72% and EPA, C20:5 ω 3 was present in minimal amounts (0.12%). This highlights the considerable seasonal variation in PUFA composition, with the current study demonstrating a more balanced $\Sigma\omega$ 3/ $\Sigma\omega$ 6 ratio in winter (0.70) compared to a decline during warmer months. Cakmak *et al.* (2012) documented a notably higher ω 3/ ω 6 ratio (2.02) in spring.

3.6. Seasonal variation in the fatty acid composition of *Cyprinus carpio*

Table 7 illustrates the seasonal variation in fatty acids of *C. carpio*. In the current study, the seasonal variation in the fatty acid composition of *C. carpio* revealed distinct patterns across seasons. For instance, Σ SFA peaked in autumn (53.01%) and winter (45.65%), largely driven by C16:0, which remained consistent at around 26% across winter, spring, and autumn. Interestingly, short-chain fatty acids such as C10:0 and C12:0 were only detected in autumn at 1.39% and 2.28%, respectively. C18:0 showed significant seasonal variation, with the highest concentration in winter (16.94%), decreasing in spring (8.54%), and slightly increasing in summer and autumn. This is consistent with the findings of Linhartova *et al.* (2018), where C16:0 and C18:0 were also prominent, with C16:0 comprising 21.33% of the total fatty acids, albeit slightly lower compared to the current study.

In the current study, Σ MUFA were the most abundant in spring (34.30%), with C18:1 ω 9 being the major contributor, peaking at 18.53% in spring and stabilizing in autumn (18.00%). This is comparable to Linhartova *et al.* (2018), who reported that C18:1 ω 9 was the most abundant MUFA in *C. carpio* (39.24%), although the concentrations in this study are notably lower. In the study on fatty acid compositions of some freshwater fish living in Porsuk Dam, Turkey, the fatty acid composition of *C. carpio* was found to include C16:0 at 21.77%, C18:0 at 6.94%, C16:1 ω 7 at 7.98%, C18:1 ω 9 at 23.61%, C18:2 ω 6 at 5.07%, C20:5 ω 3 at 5.32% and C22:6 ω 3 at 9.64% (Donmez, 2009). The study by Guler *et al.* (2008) also supports this seasonal trend, with C18:1 ω 9 fluctuating between 15.1% in spring and peaking at 20.3% in autumn.

In the current study, Σ PUFA were the highest in summer (41.16%), with C18:2 ω 6 (12.25%) and EPA, C20:5 ω 3 (11.86%) driving this increase. In contrast, PUFA levels dropped significantly in autumn (20.89%). This pattern aligns with Guler *et al.* (2008) who found that total PUFA was the highest in summer (42.8%) and lowest in winter (29.3%). The seasonal shift in the ω 3/ ω 6 ratio remained relatively stable in the current study, ranging from 0.83 in winter to 0.95 in spring, which contrasts with the findings of Guler *et al.* (2008) where the ω 3/ ω 6 ratio peaked in winter (1.06) and dropped to its lowest in autumn (0.50). Interestingly, it has been reported that *C. carpio* contained very different C20:5 ω 3 levels in two different dam lakes (Işıklı and Karacaören Dam lakes 0.56% and 20.91%, respectively) (Citil *et al.*, 2014). Additionally, the summer EPA C20:5 ω 3 (11.86%) levels observed in the current study are higher than those reported by Ljubojevic *et al.* (2013) for *C. carpio* from the Danube River (2.93%). The study by Öksüz *et al.* (2019) further highlights similar seasonal variations. They reported that C16:0 was the dominant SFA at 19.44% in winter, which is consistent with the levels found in this study. However, the PUFA composition in the current study differs, with higher values in summer (41.16%), whereas Öksüz *et al.* (2019) found total PUFA at 36.6%. In comparison to my previous study (Ekin, 2023), the current findings align with the general dominance of SFAs in *C. carpio* from the Tigris River, where SFAs made up 56.76% in summer, driven by high C16:0 levels (35.65%). However, the PUFA content in the current study is notably higher, especially in summer (41.16%) compared to the previous study (9.09%) (Ekin, 2023), indicating a significant seasonal influence on PUFA concentrations. The $\Sigma\omega$ 3/ $\Sigma\omega$ 6 ratio also showed greater stability in the current study, whereas in the previous study, this ratio was 0.08 (Ekin, 2023), indicating a higher relative proportion of ω 6 PUFAs. Overall, the comparison across studies emphasizes the significant impact of both seasonality and habitat on the fatty acid composition of *C. carpio*. The current study's findings highlight the importance of seasonal monitoring to capture the dynamic nature of lipid profiles in fish species, which are influenced by environmental factors and feeding habits (Norrbín *et al.*, 1990).

TABLE 7. Seasonal fatty acid composition of the total lipids from the dorsal muscle of *Cyprinus carpio* (% of total fatty acids)

Fatty Acids (%)	Winter	Spring	Summer	Autumn
C4:0	nd	nd	nd	nd
C6:0	nd	nd	nd	nd
C8:0	nd	nd	nd	nd
C10:0	nd	nd	nd	1.39±0.04
C11:0	nd	nd	nd	nd
C12:0	nd	nd	nd	2.28±0.04
C13:0	nd	nd	nd	nd
C14:0	2.15±0.32a	3.43±0.02b	2.60±0.20a	9.21±0.12c
C15:0	nd	0.79±0.02a	0.63±0.02a	1.06±0.02a
C16:0	26.56±1.50a	26.10±0.22a	19.92±0.22b	26.36±0.41a
C17:0	nd	1.36±0.02a	1.01±0.07a	1.00±0.01a
C18:0	16.94±0.40a	8.54±0.04b	10.12±0.02c	11.25±0.14c
C20:0	nd	nd	nd	nd
C21:0	nd	0.37±0.02	nd	nd
C22:0	nd	0.71±0.01a	nd	0.46±0.02a
C23:0	nd	nd	nd	nd
C24:0	nd	nd	nd	nd
C14:1ω5	nd	0.40±0.01a	0.65±0.02a	0.76±0.01a
C15:1ω5	nd	nd	nd	nd
C16:1ω7	2.95±0.10a	13.59±0.11b	5.39±0.20c	6.14±0.09d
C17:1ω7	nd	1.25±0.03a	0.70±0.05a	0.80±0.02a
C18:1ω9- <i>trans</i>	nd	nd	nd	nd
C18:1ω9	8.75±0.54a	18.53±0.13b	16.62±0.40c	18.00±0.20b
C20:1ω9	8.71±0.15a	0.53±0.11b	1.25±0.02b	0.82±0.01b
C22:1ω9	nd	nd	nd	nd
C24:1ω9	nd	nd	nd	nd
C18:2ω6- <i>trans</i>	nd	nd	nd	nd
C18:2ω6	10.93±1.58a	7.05±0.05b	12.25±0.05c	5.60±0.09d
C18:3ω6	6.93±0.89a	2.84±0.03b	7.63±0.39a	3.08±0.03b
C18:3ω3	7.05±0.46a	1.70±0.03b	4.28±0.17c	3.33±0.03d
C20:2ω6	nd	1.33±0.02a	nd	0.88±0.01a
C20:3ω6	0.10±0.14a	0.71±0.01a	nd	0.46±0.02a
C20:3ω3	0.07±0.10a	2.84±0.05b	nd	1.55±0.02b
C20:4ω6	0.53±0.75a	0.46±0.02a	1.62±0.07b	1.37±0.06b
C20:5ω3	5.17±0.67a	5.48±0.04a	11.86±0.02b	2.06±0.02c
C22:2ω6	nd	nd	nd	nd
C22:6ω3	3.13±0.67a	1.72±0.04b	3.52±0.20a	2.56±0.07a
ΣSFA	45.65±1.41a	41.30±0.25b	34.28±0.31c	53.01±0.63d
ΣMUFA	20.41±0.70a	34.30±0.17b	24.61±0.24c	26.52±0.27d
ΣPUFA	33.91±1.40a	24.13±0.10b	41.16±0.31c	20.89±0.13d
Σω6	18.49±0.53a	12.39±0.04b	21.50±0.10c	11.39±0.08b
Σω3	15.42±0.87a	11.74±0.06b	19.66±0.24c	9.50±0.05d
Σω3/Σω6	0.83	0.95	0.91	0.83

Values are expressed as means ± SD for three replicates and are reported as percentages of total fatty acid methyl esters. The mean values of different characters within the same row were compared using one-way ANOVA followed by Tukey's post-hoc test. Different letters indicate significant differences ($P \leq 0.05$). Results are expressed as % fatty acids of total lipids. nd – not detected.

So far, the above result and discussion section has compared studies examining the same fish species. In different freshwater fish taxa, the dominant fatty acids are also C16:0, C16:1 ω 7, and C18:1 ω 9, with moderate levels of C14:0 and C18:0. Functional fatty acids such as C18:2 ω 6, C18:3 ω 3, C20:4 ω 6, C20:5 ω 3, and C22:6 ω 3 are also significant components, though their proportions vary by species and environmental factors. For example, in *Salmo trutta labrax*, C16:0 (25.39%), C16:1 ω 7 (5.63%), C18:0 (5.91%), C18:1 ω 9 (20.63%), and C22:6 ω 3 (21.42%) were predominant (Aras, 2003). Similarly, in *Oncorhynchus mykiss*, C16:0 (21.3%), C16:1 ω 7 (4.16%), C18:0 (6.79%), C18:1 ω 9 (22.2%), and C22:6 ω 3 (22.7%) were reported as major components (Haliloğlu *et al.*, 2004). Studies on species from Turkey, including *Barbus lacerta*, *Squalius berak*, *Chondrostoma regium*, and *Silurus triostegus*, revealed a consistent dominance of C16:0, C18:0, C16:1 ω 7, and C18:1 ω 9 (Cengiz *et al.*, 2010; Ekin, 2023). Other findings from *Scardinius erythrophthalmus* and *Anguilla anguilla* in the Büyük Menderes River (Bayar *et al.*, 2021) and from *Carassius carassius*, *Leuciscus cephalus*, and *Tinca tinca* in Porsuk Dam (Donmez, 2009) confirmed similar patterns. However, while high levels of C20:4 ω 6, C20:5 ω 3, and C22:6 ω 3 were reported in *C. regium*, *B. rajonorum*, *Leuciscus lepidus*, and *Acanthobrama marmid* (Cengiz *et al.*, 2010), some studies found these components below 1% in freshwater species (Citil *et al.*, 2014; Kaçar *et al.*, 2018; Łuczyńska *et al.*, 2014).

4. CONCLUSIONS

C. gibelio exhibited the highest total lipid content, ranging from 7.57% in winter to 8.55% in summer and 8.30% in autumn. *C. trutta* and *C. umbla* reached their Σ PUFA maxima in winter at 38.31% and 43.06%, respectively, driven by EPA (12.21% in *C. trutta*; 9.27% in *C. umbla*) and linoleic acid (C18:2 ω 6) (10.85% vs. 19.14%). *C. luteus* showed exceptionally high DHA (C22:6 ω 3) levels, peaking at 16.69% in summer, alongside Σ PUFA of 48.30% in summer and 48.88% in autumn. *C. gibelio* also had elevated Σ PUFA values (49.89% in winter, 49.33% in summer) and achieved the most favorable ω 3/ ω 6 ratio of 2.04 in spring. Interspecific and seasonal variations highlight the importance of selecting species and timing consumption to maximize health benefits: *C. gibelio* for high lipid diets, *C. trutta* and *C. umbla* in winter for EPA/linoleic acid, and *C. luteus* in summer–autumn for DHA. Functional fatty acids such as C16:0 and C18:1 ω 9 were consistently dominant, aligning with regional data on freshwater taxa. These findings provide critical insights for the sustainable management of fisheries and public health strategies. Given their widespread distribution across Mesopotamia, Anatolia, and the Balkans, these fish contribute significantly to traditional diets and food security.

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DECLARATION OF COMPETING INTEREST

The authors declare that there are no conflicts of interest or financial disclosures related to this study.

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AUTHORSHIP CONTRIBUTION STATEMENT

All aspects of this study, including conceptualization, data collection, analysis, and manuscript writing, were conducted by İE. The laboratory analyses were carried out as part of a project at a food laboratory in Istanbul.

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