

# Determination of fatty acids and evaluation of nutritional quality of *Barbus lacerta* from Murat River, Türkiye

## Murat Nehri, Türkiye'den *Barbus lacerta*'nın yağ asitlerinin belirlenmesi ve besin kalitesinin değerlendirilmesi

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**Abstract:** The ultimate goal, to investigate for differences in seasonally fatty acid composition of liver, muscle and gonad from *Barbus lacerta* females' fish, bought from fishermen who hunt from the Murat River, but also to reveal the food quality in edible muscle tissue with nutritional quality index fatty acids. Fatty acid composition of *B. lacerta* varied different among the three tissues/organs and the most important difference was seen between muscle and liver. *B. lacerta* had found higher proportions in 14:0, 16:0, EPA and DHA in muscle and liver than the gonads. In contrast, the gonads had higher proportions of ARA because of the important role of ARA in the ovulation processes (late winter-early spring). In parallel, there were changes in the food quality indexes (AI, TI, h/H, DFA, OFA,  $\omega 6/\omega 3$ , DHA+EPA, HPI, PUFA/SFA) used based on fatty acids in determining the food quality in edible muscle tissue. The highest change was in AI (Atherogenicity index), although there were no significant seasonal differences. The lowest AI values as good quality index was determined in spring (0.42) and winter (0.41). The food quality index that stood out in each season was different. Therefore, it can be suggested that *B. lacerta* can be consumed as a high quality and healthy food, partially except for summer.

**Keywords:** Atherogenicity index, arachidonic acid, *Barbus lacerta*, food quality

**Öz:** Murat Nehri'nde avlanan balıkçılardan satın alınan dişi *B. lacerta* balıklarının karaciğer, kas ve gonadlarındaki mevsimsel yağ asidi bileşimlerindeki farklılıkları araştırmak ve ayrıca besin kalitesi indeksi yağ asitleri ile yenilebilir kas dokusundaki besin kalitesinin ortaya konulması amaçlanmıştır. *B. lacerta*'nın yağ asidi bileşimi üç doku/organ arasında farklılık göstermiştir ve en önemli fark kas ve karaciğer arasında görülmüştür. *B. lacerta*, kas ve karaciğerlerinde gonadlarından daha yüksek oranlarda 14:0, 16:0, EPA ve DHA bulmuştur. Buna karşılık, ARA'nın yumurtlama süreçlerindeki (kış sonu-ilkbahar başı) önemli rolü nedeniyle gonadlar daha yüksek ARA oranlarına sahipti. Buna paralel olarak, yenilebilir kas dokusundaki besin kalitesini belirlemede yağ asitlerine dayalı olarak kullanılan besin kalitesi indekslerinde (AI, TI, h/H, DFA, OFA,  $\omega 6/\omega 3$ , DHA+EPA, HPI, PUFA/SFA) değişiklikler olmuştur. En yüksek değişim AI (Aterojenite indeksi) da olmakla birlikte önemli mevsimsel farklılıklar yoktu. Besin kalite indeksi olarak en düşük AI değerleri ilkbaharda (0.42) ve kışın (0.41) belirlendi. Her mevsimde öne çıkan gıda kalite indeksi farklıydı. Bu nedenle, *B. lacerta*'nın yüksek kaliteli ve sağlıklı bir gıda olarak tüketimi kısmen yaz hariç önerilebilir.

**Anahtar kelimeler:** Aterojenite indeksi, araşidonik asit, *Barbus lacerta*, gıda kalitesi

## INTRODUCTION

Fatty acids as dietary nutrients are important compounds as energy-rich biochemical for consumers and can be used as tracers of organic matter pathways in aquatic food webs. Aquatic ecosystems are the primary source of omega-3 fatty acids ( $\omega 3$  FAs), because of supporting both aquatic and terrestrial heterotrophs by the trophic transfer of these key essential fatty acids (EFAs) via food webs (Gladyshev et al., 2013). It is known that fish meat is a valuable food in human nutrition (Ahmed et al., 2022). A large body of research has detailed how the growth and reproduction of many aquatic consumers can be restricted by the limitation of EFA, suggesting that fatty acids are a promising biochemical metric for use as a proxy for ecosystem-scale food quality (Galloway and Winder 2015). In nature, fatty acids are found in the form of mixtures of saturated fatty acids (SFAs), monounsaturated fatty acids (MUFAs) and polyunsaturated fatty acids (PUFAs). Especially, PUFA are vital dietary elements that play an important role in human nutrition. In addition to providing the

body with an important source of energy such as SFA, PUFA obtained from fish play a role in cell membrane structure, blood pressure regulation, and coagulation; participate in the proper functioning of the immune system and the assimilation of fat-soluble vitamins; affect the synthesis of pro- and anti-inflammatory substances; and protect the cardiovascular system (Mititelu et al., 2024).

Therefore, their nutritional and medicinal values should be determined. In recent years, classical indices such as SFA, MUFA, PUFA,  $\omega 6$ ,  $\omega 3$  and  $\omega 6/\omega 3$  have not been taken into account much. Instead, indices such as PUFA/SFA, atherogenicity index (AI), thrombogenicity index (TI), hypocholesterolemic/hypercholesterolemic ratio (h/H), health-promoting index (HPI) and sum of eicosapentaenoic acid and docosahexaenoic acid (EPA + DHA) are taken into account for fish species (Chen and Liu, 2020).

FA composition of different tissues and organs may vary

according to their specific physiological roles (Parzanini et al., 2021). Muscle and liver are the main tissues that mainly consist of triacylglycerols (TAG) and storage fats (Pierron et al., 2009). The liver is the main site of lipogenesis (Tocher, 2003) and energy conversion (Hansen and Abraham, 1983) in fish. Gonads are more resistant to changes in fatty acid composition when compared to other tissues, since its composition is genetically determined to allow adequate embryo development (Sargent et al., 2002). For example, ARA (Arachidonic acid, 20:4 $\omega$ 6) is one of the major nutrients to ensure reproductive success in many fish species (Tocher, 2010), as is the main precursor for the 2-series prostaglandins (PG-2), eicosanoids that stimulate steroid synthesis in the ovary, trigger oocyte maturation and affect the sexual behavior of females (Tocher, 2003).

Although there are many studies on seasonal changes in many different freshwater fish species and some *Barbus* species, no reports have yet been published on the effects of seasonal changes on the fatty acid composition and nutritional quality with food quality index fatty acids of the freshwater female Kura Barbell (*Barbus lacerta*, Heckel 1843). *Barbus lacerta* is an Asian origin and distributed in the Euphrates-Tigris rivers in the Mesopotamian basin. *B. lacerta* is a member of Cyprinidae family from ray-finned fishes. It is an endemic fish species for inland waters of Eastern Anatolia, Türkiye (Geldiay and Balık, 2007). Based on all these, it was aimed to evaluate the fatty acid composition of *B. lacerta* both as an ecological and nutritional source. We put forward the hypothesis that these changes will show significant differences in different tissues seasonally and this may have important effects on the nutritional quality of edible muscle tissue. We think that this will shed light on the hunting and consumption of this fish species during the period when the nutritional quality is best. Tissues (muscle, liver and gonad) of female *B. lacerta* was used in the study because females had higher length and weight than males. (Dopeikar et al., 2015; Şen Özdemir et al., 2023). All these increase the economic value of female *B. lacerta*. For these reasons, we preferred to examine the differences in fatty acid composition among the three tissues of females.

## MATERIALS AND METHODS

### Preliminary preparations

*Barbus lacerta* samples were obtained from contracted commercial fishermen. Samplings were done randomly in different months to represent the seasons from different points in Göynük Stream, Murat River (Türkiye). Nets with different eye apertures were used in catching the fishes. The catching fish were brought to the laboratory on the ice (September 2018-August 2019). Sexually mature fish were used in the analysis. Fish were cut according to the butterfly fillet technique (Liu et al., 2022). The long-based fins, head and skin were cut off. The meat along the "top" of the fish was cut and separated from the bones. The backbone was separated from the tail and lift the spine from the tail and pull out the meat, removing a butterfly fillet from the fish. The muscle tissues of fish were separated from the inedible parts of the deceased fish. The internal organs (liver and gonad) were removed by pliers. The gender

determination was made macroscopically from the gonads of the fish catching fish. The study was carried out on the adult female fish (mean total weight=32 kg; mean total length=15 cm) (32-53 individuals). Then, every tissue/organ sample was sealed in plastic bags. All the fish muscle were stored at -80 °C for further analysis.

### Lipid extraction and fatty acid derivatization

Lipid was extracted from muscle, liver and gonad tissues separated from caught fish. Hara and Radin (1978) method were used in hexane: isopropanol (3:2) solution for lipid extraction. These tissues are separately taken from each sampling period and then centrifuged at 4000 rpm for 10 min, leading to the formation of two separate layers. The supernatant layer was removed from the organic layer and transferred into new tubes. 5 ml 2% methanolic sulfuric acid solution was added to the supernatant layer and mixed completely with the vortex. The mixture was left to be methylated in an oven at 55 °C for 15 hours. Then, 5 ml of 5% NaCl was added and mixed. 5 ml of hexane was added to the tubes and waited about 3 hours at room temperature. The hexane phase formed was taken from the top, 5 ml 2% KHCO<sub>3</sub> solution was added to the tubes. The final extracts were evaporated using a gentle stream of nitrogen evaporator (Allsheng WD-12). Then, 1 ml hexane was added to the dry lipid layer and the mixture was vortexed. Every sample was taken into 2 ml labelled autosampler vials and waited -20 °C until mass spectrometer gas chromatograph (GC/MS) analysis.

### GC/MS analysis

GC/MS (American, Agilent 5975 C) was used for the FAME analysis. Machery-Nagel (Germany) capillary column (30 m x 0.25 mm, 0.25  $\mu$ m) was used. The column temperature was 120-220 °C, the injection temperature was 240 °C and the detector temperature was 280 °C. Helium (He) (0.5 ml/min) was used as a carrier gas (David et al., 2005). FAME of the catching fish were made from the retention times of each fatty acid using standard (Supelco: 37 component FAME mix (Product number 47885-U). After analysis, wsearch32 software was used in the identification of the peaks of each fatty acid. Individual FA data were reported as percentage weights (%) of total identified FA.

### Determination of nutritional quality of *B. lacerta*

There is certain fatty acid groups used as nutritional quality index in fish. In this study, the following 9 indices were used to determine the nutritional quality index of *B. lacerta*.

#### 1. Atherogenicity index (AI)

AI= [12:0 + (4 $\times$ 14:0) + 16:0]/( $\omega$ 3PUFA +  $\omega$ 6PUFA + MUFA)] (Ulbricht and Southgate 1991; Garaffo et al., 2011; Luczyńska and Paszczyk 2019)

#### 2. Thrombogenicity Index (TI)

TI= [14:0 + 16:0 + 18:0]/[(0.5 $\times$ 18:1) + (0.5 $\times$  $\Sigma$ MUFA) + (0.5 $\times$  $\Sigma$ PUFA $\omega$ 6) + (3 $\times$  $\Sigma$ PUFA $\omega$ 3) +  $\Sigma$ PUFA $\omega$ 3/ $\Sigma$ PUFA $\omega$ 6)] (Ulbricht and Southgate 1991; Santos-Silva et al., 2002; Garaffo et al., 2011; Luczyńska and Paszczyk 2019)

## 3. Hypocholesterolemic/hypercholesterolemic ratio (h/H)

$$h/H = [(18:1 + 18:2 + 18:3 + 20:3 + 20:4 + 20:5 + 22:4 + 22:5 + 22:6) / (14:0 + 16:0)]$$
 (Santos-Silva et al., 2002)

## 4. Hypercholesterolaemic fatty acids (OFAs)

$$OFA = 12:0 + 14:0 + 16:0$$

High OFA content reduces the lipid quality of food. (Łuczyńska and Paszczyk 2019)

## 5. Polyunsaturated fatty acids/saturated fatty acids ratio (PUFA/SFA)

## 6. Total of eicosapentaenoic acid and docosahexaenoic acid (EPA+DHA)

7. Polyunsaturated Omega 6 Fatty Acids/ Polyunsaturated Omega 3 Fatty Acids ( $\omega 6/\omega 3$ )

## 8. Desirable fatty acids (DFAs)

$$DFA = 18:0 + \Sigma MUFA + \Sigma PUFA$$
 (Costa et al., 2008; Silva et al., 2019; Łuczyńska and Paszczyk 2019; Tibaoui et al., 2020).

## 9. The health-promoting index (HPI)

$$HPI = \Sigma MUFA + \Sigma PUFA / [12:0 + (4 \times 14:0) + 16:0]$$
 (Chen and Liu 2020).

## Statistical analysis

Only individual FAs present with mean proportions  $\geq 0.5\%$  across the different tissues/organs (muscle, liver and gonad) were analyzed statistically. Multivariate statistics were used to analyze differences in fatty acid composition in PRIMER-e 2017. The Bray Curtis similarity coefficient was used for PERMANOVA, principal coordinates (PCO) and CLUSTER analysis for similarity ranges. In analyses of the entire fatty acid data's (total fatty acids, food quality index fatty acids) of *B. lacerta* factored by season, the tissues/organs (muscle, liver, gonad). PERMANOVA tests and SIMPER (Cut off for low contributions: 70%) were conducted on Bray–Curtis similarities with the unrestricted permutation of raw data method (number of permutations 9999; type III sums of squares). The tests were used to identify the fatty acids that contributed the most to the similarities between/within the factor groups.

To further investigate the seasonal effects on changes in nutritional quality in edible muscle tissue and differences in fatty acid composition among tissues/organs, one-way analysis of variance (ANOVA) (Post-Hoc, Homogeneous Groups, Significant Differences) was conducted. ANOVA tested the significance ( $p < 0.05$ ) of the differences in the effect of seasons and tissues/organs (liver, muscle, gonad) on fatty acids using STATISTICA software.

## RESULTS

Fatty acid composition of *B. lacerta* tissues/organs (muscle, liver, gonad)

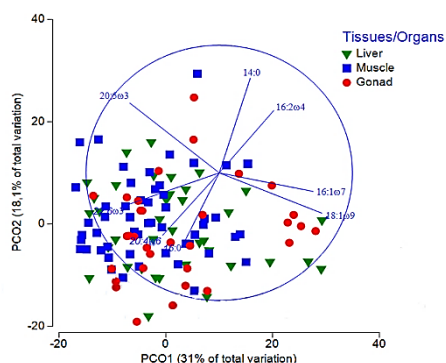
The fatty acid composition in all *B. lacerta* tissues/organs taken during the sampling period was investigated regardless of the sampling season. FA composition varied differently among the three tissues/organs (PERMANOVA, Pseudo-

$F = 3.48$ ,  $P(\text{perm}) = 0.0002$ ) (Figure 1). The most important difference was between the muscle and liver, muscle and gonad with the same  $P(\text{perm})$  value = 0.002. However, the difference between muscle and liver was higher ( $t = 1.99$ ) than the other ( $t = 1.97$ ). The average similarity within tissues/organs changed between 71–76 % (liver-muscle) according to SIMPER results. The fatty acids that contribute the most to the similarity were 16:0 (20%; 22%, respectively) within the liver and gonads. EPA was the first contributor within muscle with 21%. However, EPA was in the second contributor within liver and gonad with the same value 17%. The highest difference between liver and gonad was in ARA from PUFA, between liver and muscle in  $\omega 16:0$  from SFA (Figure 2, Table 1). ARA was the lowest in the liver, the highest in the gonad, while the EPA and DHA were lower in the gonad and the highest EPA was in the liver, the lowest DHA was in the muscle tissue. However, the percentage of LC-PUFA in the muscle and liver of *B. lacerta* was higher than gonad (Table 1). The highest difference between liver and gonad was in ARA from PUFA, between liver and muscle in  $\omega 16:0$  from SFA. There was no difference between the tissues/organs in  $\Sigma$ SFA, while there was a difference between the tissues/organs in  $\Sigma$ MUFA,  $\Sigma$ PUFA and  $\omega 3$  fatty acids, (Table 1,  $p < 0.05$ ).

**Table 1.** Mean fatty acid composition for muscle, liver and gonad in catching fish of female *B. lacerta* during the sampling period (% of total FA)

FAs	LIVER (n=39)	MUSCLE (n=53)	GONAD (Ovary) (n=32)
12:0	1.07±0.99	1.10±0.98	1.15±1.11
14:0	2.91±2.07	2.78±1.80	3.30±2.21
15:0	0.54±0.40	0.45±0.27	0.54±0.48
16:0	0.58±0.57 <sup>b</sup>	1.36±1.00 <sup>a</sup>	1.13±0.92 <sup>a</sup>
16:1	16.71±4.18	17.70±5.32	18.37±4.31
17:0	1.30±1.08	0.81±0.53	0.70±0.58
18:0	3.88±2.18	3.22±2.01	4.04±2.45
20:0	0.76±0.56	0.54±0.41	0.73±0.52
$\Sigma$ SFA	28.30±6.01	28.57±4.94	30.43±5.90
MSFA*	0.55±0.49 <sup>a</sup>	0.61±0.78 <sup>a</sup>	0.47±0.22 <sup>b</sup>
14:1	0.73±0.68	0.67±0.50	0.84±0.51
16:1 $\omega$ 7	13.87±5.52 <sup>a</sup>	11.52±3.37 <sup>b</sup>	13.85±4.42 <sup>a</sup>
17:1	0.62±0.47	-	-
18:1 $\omega$ 9	10.86±6.78	8.90±4.81	10.39±6.31
20:1 $\omega$ 9	2.28±2.03 <sup>a</sup>	1.37±1.07 <sup>b</sup>	1.10±0.79 <sup>b</sup>
22:1 $\omega$ 11	-	-	0.62±0.55
22:1 $\omega$ 9	0.59±0.45	-	-
22:1 $\omega$ 7	-	-	0.62±0.42
$\Sigma$ MUFA	29.25±10.12 <sup>a</sup>	24.03±6.55 <sup>b</sup>	28.33±10.36 <sup>ab</sup>
MMUFA*	0.30±0.26 <sup>c</sup>	1.57±0.86 <sup>a</sup>	0.91±0.62 <sup>b</sup>
16:2 $\omega$ 4	0.52±0.47	1.26±0.74	1.09±0.58
18:2 $\omega$ 6	2.64±2.32	2.76±2.01	2.08±1.49
18:3 $\omega$ 3	3.39±2.03	4.47±2.31	3.79±2.87
20:4 $\omega$ 6 (ARA)	4.55±2.78 <sup>b</sup>	5.70±2.22 <sup>b</sup>	7.46±3.37 <sup>a</sup>
20:5 $\omega$ 3 (EPA)	15.11±5.78 <sup>b</sup>	18.76±4.66 <sup>a</sup>	14.81±5.14 <sup>b</sup>
22:2	1.28±0.77	1.36±0.59	1.13±0.38
22:6 $\omega$ 3 (DHA)	14.05±6.26 <sup>a</sup>	12.56±4.94 <sup>ab</sup>	10.00±5.40 <sup>b</sup>
$\Sigma$ PUFA	42.45±9.07 <sup>b</sup>	47.40±7.08 <sup>a</sup>	41.32±8.79 <sup>b</sup>
MPUFA*	0.91±0.82 <sup>a</sup>	0.53±0.42 <sup>b</sup>	0.96±0.69 <sup>a</sup>

\*Minor FA < 0.5 (19:0, 21:0, 23:0 from SFA; 15:1, 20:1 $\omega$ 7, 20:1 $\omega$ 11 from MUFA; 18:3 $\omega$ 4, 18:3 $\omega$ 6, 20:2 $\omega$ 6, 22:5 $\omega$ 3 from PUFA. Means followed by different letters (a, b, c), letter groups in the same row are significantly different ( $p < 0.05$ ), while means do not differ if there are no letters in the same row ( $p < 0.05$ ). ± SD (Standard Deviation), '-' non defined.



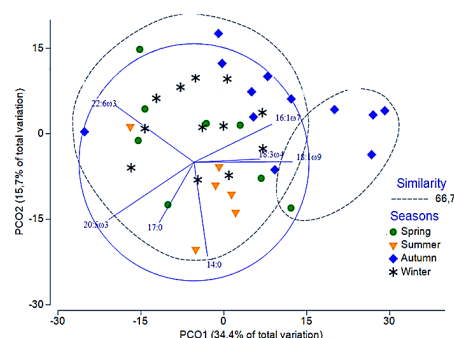
**Figure 1.** Differences in the fatty acid composition of *B. lacerta* tissues/organs plotted using PCO. The lower triangular matrix was created using Bray–Curtis similarity coefficients. Pearson correlation ( $p>0.60$ ).

### Seasonally FA composition of *B. lacerta* tissues/organs (muscle, liver, gonad)

#### Liver

The FA composition of *B. lacerta* liver varied across seasons (PERMANOVA, Pseudo- $F=2.79$ ,  $P$  (perm)=0.0003, Pearson Correlation  $p>0.55$ ). The most seasonal differences were between summer and autumn ( $t=1.97$   $P$  (perm)=0.004). The most average similarity was within winter (77%). The fatty acids that contributed the most to this similarity were 16:0 (palmitic acid) (21%), EPA (19%), 16:1 $\omega$ 7 (15%), DHA (14%) and 18:1 $\omega$ 9 (8%) (Bray-Curtis Similarity, Cut off for low

contributions: 70 %). Seasonally fatty acid composition of liver was important for 14:0, 18:0, 18: $\omega$ 9, EPA,  $\omega$ 3 fatty acids. 14:0, ARA, EPA and DHA were characteristic fatty acids for all seasons in liver. However, 16:1 $\omega$ 7, 18:1 $\omega$ 9 and 18:3 $\omega$ 4 were more characteristic fatty acids for autumn than other seasons with similarity 66.7% (Figure 2). The most important difference was only  $\Sigma$ MUFA in total FA groups (Table 2), especially between autumn and winter ( $p<0.05$ ). The most significant difference was between summer and winter for 14:0 autumn and winter for 16:1 $\omega$ 7 and following  $\omega$ 3 fatty acids between spring and autumn. The difference between autumn and winter, where 18:1 $\omega$ 9 differed, was partially significant



**Figure 2.** Two-dimensional configuration plot of a PCO analysis of a resemblance matrix of fatty acids in liver of *B. lacerta*. The lower triangular matrix was created using Bray–Curtis similarity coefficients. Pearson Correlation ( $p>0.55$ )

**Table 2.** Seasonally fatty acid composition for liver of *B. lacerta* (% of total FA)

FA	LIVER			
	Spring (n=9)	Summer (n=6)	Autumn (n=12)	Winter (n=12)
12:0	1.19±0.99	1.29±0.62	0.55±0.38	1.43±0.34
14:0	3.16±1.51 <sup>ab</sup>	5.50±3.13 <sup>a</sup>	2.26±0.66 <sup>b</sup>	2.15±0.86 <sup>b</sup>
16:0	-	-	0.62±0.23 <sup>b</sup>	0.94±0.45 <sup>a</sup>
15:0	0.61±0.32	0.51±0.25	0.53±0.31	0.50±0.28
16:0	15.46±4.24	14.72±4.83	17.15±6.79	18.20±4.04
17:0	2.67±1.56 <sup>a</sup>	1.52±0.12 <sup>ab</sup>	-	1.01±0.23 <sup>b</sup>
18:0	3.02±1.02 <sup>b</sup>	6.53±3.18 <sup>a</sup>	3.69±1.58 <sup>b</sup>	3.63±1.99 <sup>b</sup>
20:0	1.17±0.34	1.27±0.56	-	0.60±0.45
23:0	-	0.67±0.48	-	-
*MSFA	0.84±0.76 <sup>a</sup>	0.56±0.45 <sup>b</sup>	0.60±0.48 <sup>ab</sup>	0.46±0.65 <sup>b</sup>
$\Sigma$ SFA	28.12±4.12	32.57±4.78	25.40±3.12	28.92±2.78
14:1	0.50±0.30	1.35±1.03	0.51±0.25	0.80±0.42
16:1 $\omega$ 7	13.72±3.12	9.06±3.25	16.19±7.10	14.06±4.44
17:1	0.94±0.22	0.86±0.31	-	-
18:1 $\omega$ 9	9.35±5.22 <sup>ab</sup>	9.54±2.70 <sup>ab</sup>	15.66±8.44 <sup>a</sup>	8.38±3.84 <sup>b</sup>
20:1 $\omega$ 9	1.39±0.98	2.84±2.12	2.55±2.10	2.02±1.40
22:1 $\omega$ 9	-	-	1.18±0.56	0.58±0.36
22:1 $\omega$ 11	-	-	-	-
*MMUFA	0.57±0.46 <sup>b</sup>	1.43±0.84 <sup>a</sup>	1.63±1.34 <sup>a</sup>	0.91±0.76 <sup>b</sup>
$\Sigma$ MUFA	26.47±7.14 <sup>b</sup>	25.08±6.23 <sup>b</sup>	37.72±8.56 <sup>a</sup>	26.75±5.67 <sup>b</sup>
16:2 $\omega$ 4	-	-	0.83±0.34	0.70±0.42
18:2 $\omega$ 6	1.41±0.85	2.70±1.53	4.56±3.22	1.77±1.02
18:3 $\omega$ 3	2.91±1.70	4.86±3.22	3.65±3.12	2.94±2.05
ARA	3.51±2.20	3.40±1.80	4.52±3.45	5.78±2.19
EPA	17.60±3.54 <sup>a</sup>	17.57±3.96 <sup>a</sup>	10.64±5.74 <sup>b</sup>	16.97±3.72 <sup>a</sup>
22:2	1.81±0.97 <sup>a</sup>	1.39±0.93 <sup>ab</sup>	0.88±0.35 <sup>b</sup>	1.22±0.62 <sup>ab</sup>
DHA	18.29±9.37	11.76±6.66	11.55±5.23	14.24±5.89
*MPUFA	0.78±0.72	0.67±0.34	0.23±0.18	0.71±0.48
$\Sigma$ PUFA	46.31±8.54	42.35±6.96	36.86±4.12	44.33±6.23
$\Sigma\omega$ 3	38.79±5.12 <sup>a</sup>	34.19±4.87 <sup>ab</sup>	25.84±3.18 <sup>b</sup>	34.15±3.89 <sup>ab</sup>
DHA/EPA	1.16±0.34	0.68±0.26	1.18±0.45	0.86±0.42

\*Minor FA<0.5 (19:0, 21:0, 23:0 from SFA; 15:1, 20:1 $\omega$ 7, 20:1 $\omega$ 11 from MUFA; 18:3 $\omega$ 4, 18:3 $\omega$ 6, 20:2 $\omega$ 6, 22:5 $\omega$ 3 from PUFA). Means followed by different letters (a,b), letter group in the same row are significantly different ( $p<0.05$ ), while means do not differ if there are no letters in the same row ( $p<0.05$ ).  $\pm$  SD (Standard Deviation), '-' non defined.



### Muscle

The FA composition of *B. lacerta* muscle varied across the seasons (PERMANOVA, pseudo- $F=2.10$ ,  $P(\text{perm})=0.008$ , Pearson correlation  $p>0.60$ ). The most seasonal differences were between spring and autumn ( $t=1.71$ ,  $P(\text{perm})=0.02$ ,  $p>0.60$ ). The most average similarity was within autumn (79 %). The FA that contributed the most to this similarity were 16:0 (21%), EPA (17%), DHA (14%), 16:1 $\omega$ 9 (13%) and 18:1 $\omega$ 9 (10%) (Bray-Curtis Similarity, Cut off for low contributions: 70%). 14:0, 12:0, 16:0, 16:1 $\omega$ 7, EPA and DHA were characteristic FA for all seasons. However, 16:0 and ARA were more characteristic FA for winter and 18:1 $\omega$ 9 for autumn than other seasons (Figure 3). No difference was in total FA groups (Table 3) ( $p<0.05$ ). The most significant difference was between spring and autumn for EPA and following 22:2 between spring and summer, 16:0 between summer and winter (Table 2,  $p<0.05$ ).

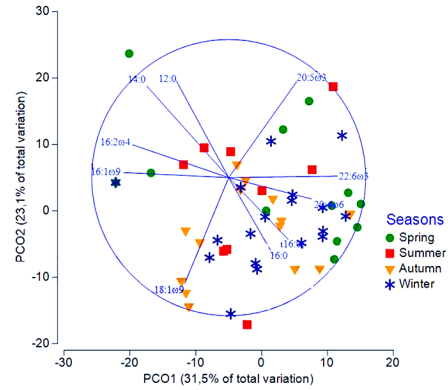


Figure 3. Differences in seasonally fatty acid composition of *B. lacerta* muscle plotted using PCO. The lower triangular matrix was created using Bray-Curtis similarity coefficients. Pearson correlation ( $p>0.60$ ).

Table 3. Seasonally fatty acid composition for muscle of *B. lacerta* (% of total FA)

FA	MUSCLE			
	Spring (n=12)	Summer (n=9)	Autumn (n=14)	Winter (n=18)
12:0	1.56±1.37	1.12±0.41	0.83±0.77	0.99±0.70
14:0	3.51±1.15 <sup>ab</sup>	3.90±1.48 <sup>a</sup>	2.44±1.06 <sup>ab</sup>	2.00±1.15 <sup>b</sup>
16:0	1.32±0.36 <sup>ab</sup>	0.71±0.41 <sup>b</sup>	1.24±0.47 <sup>ab</sup>	1.81±0.51 <sup>a</sup>
15:0	-	0.63±0.45	-	-
16:0	14.61±3.90	19.38±4.74	18.42±5.95	18.38±2.58
17:0	1.26±0.56	0.93±0.45	-	0.75±0.38
18:0	2.74±2.01	3.18±1.71	3.97±2.06	2.74±1.44
20:0	0.80±0.58	0.67±0.35	-	-
23:0	-	-	-	0.77±0.46
*MSFA	3.00±2.30 <sup>a</sup>	0.56±0.41 <sup>b</sup>	1.79±0.88 <sup>b</sup>	0.97±2.26 <sup>ab</sup>
ΣSFA	28.80±3.17	31.08±0.41	28.69±5.85	28.41±3.88
14:1	0.51±0.32	0.98±0.67	0.55±0.34	0.72±0.41
16:1 $\omega$ 7	10.42±4.34	12.78±1.32	12.05±2.77	11.21±3.15
17:1	0.60±0.52	-	-	-
18:1 $\omega$ 9	8.42±3.44	6.72±4.82	10.77±6.04	8.85±4.84
20:1 $\omega$ 9	1.91±1.43	0.96±0.77	1.19±1.12	1.37±0.79
22:1 $\omega$ 9	-	-	-	-
22:1 $\omega$ 11	-	-	-	0.78±0.36
*MUFA	0.73±0.59	1.08±0.52	1.78±1.52	1.02±0.96
ΣMUFA	22.59±5.92	22.52±4.32	26.34±8.14	23.95±7.19
16:2 $\omega$ 4	1.72±1.05	0.89±2.19	1.26±1.23	1.15±1.21
18:2 $\omega$ 6	1.98±0.95	3.11±1.87	3.27±2.75	2.71±1.36
18:3 $\omega$ 3	3.15±2.25 <sup>b</sup>	5.86±2.00 <sup>a</sup>	5.08±1.96 <sup>ab</sup>	4.16±2.84 <sup>ab</sup>
ARA	5.40±2.46	5.07±0.59	4.95±1.56	6.81±2.25
EPA	21.12±4.54 <sup>a</sup>	18.57±3.75 <sup>ab</sup>	16.01±5.61 <sup>b</sup>	19.41±3.65 <sup>ab</sup>
22:2	1.67±0.77 <sup>a</sup>	0.95±0.25 <sup>b</sup>	1.21±0.43 <sup>ab</sup>	1.48±0.39 <sup>ab</sup>
DHA	15.12±7.21	11.46±1.70	12.66±3.68	11.34±3.04
*MPUFA	0.45±0.28	0.49±0.18	0.53±0.21	0.58±0.32
ΣPUFA	50.61±4.93	46.40±5.19	44.97±9.07	47.64±5.08
Σ $\omega$ 3	39.39±6.47	35.88±3.06	33.76±9.17	34.91±5.77
DHA/EPA	0.75±0.43	0.64±0.24	0.84±0.32	0.59±0.18

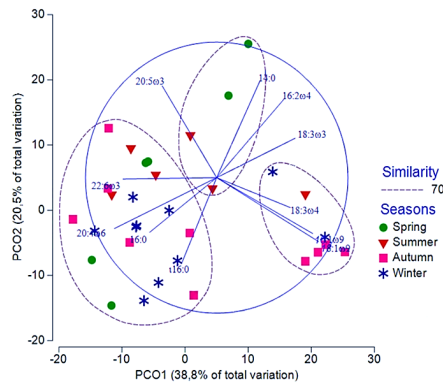
\*Minor FA<0.5 (19:0, 21:0, 23:0 from SFA; 15:1, 20:1 $\omega$ 7, 20:1 $\omega$ 11 from MUFA; 18:3 $\omega$ 4, 18:3 $\omega$ 6, 20:2 $\omega$ 6, 22:5 $\omega$ 3 from PUFA). Means followed by different letters, letter groups in the same row are significantly different ( $p<0.05$ ), while means do not differ if there are no letters in the same row ( $p<0.05$ ). ± SD (Standard Deviation), '-' non defined.

### Gonad (Ovary)

The FA composition of *B. lacerta* gonads did not vary significantly across the season (PERMANOVA, Pseudo- $F=1.30$ ,  $P(\text{perm})=0.21$ , Pearson correlation  $p>0.60$ ). The most seasonal differences were between summer and winter ( $t=1.38$ ,  $P(\text{perm})=0.09$ ). The most average similarity was within winter (77%).

The fatty acids that contributed the most to this similarity were 16:0 (21%), 16:1 $\omega$ 9 (17%), EPA (17 %), 18:1 $\omega$ 9 (11%) and DHA

(9%), and (Bray-Curtis Similarity, Cut off for low contributions: 70%). 16:0, 16:0, ARA and DHA were characteristic fatty acids for all seasons in gonads. However, 18:1 $\omega$ 9 and 16:1 $\omega$ 7 were more characteristic fatty acids for autumn and winter. Additionally, EPA, 14:0, 16:2 $\omega$ 4 were more characteristic fatty acids for summer and spring than other seasons with 70% similarity (Figure 4). No difference was in total fatty acid groups (Table 4) ( $p<0.05$ ). The most significant difference was between spring and winter for 16:0 and the following 14:0 between summer and winter, summer and autumn (Table 2,  $p<0.05$ ).



**Figure 4.** Two-dimensional configuration plot of a PCO analysis of a resemblance matrix of fatty acids in gonad of *B. lacerta*. The lower triangular matrix was created using Bray–Curtis similarity coefficients. Pearson Correlation ( $p>0.60$ ).

### Seasonally FA composition of *B. lacerta* edible muscles

The nutritional quality of *B. lacerta* edible muscle tissue did not vary significantly across the season to SIMPER results; the most average similarity was within winter (94%) for nutritional quality index fatty acids of *B. lacerta* muscle tissue. The FA that contributed the most to this similarity were DFA (56%) and EPA+DHA (19%) (Bray-Curtis Similarity, Cut off for low contributions: 70%). DFA and EPA+ DHA were the most characteristic nutritional indexes for all the seasons. The lowest dissimilarity was between spring and winter (7%), while the highest dissimilarity was between summer and autumn, spring and summer with 10%. No difference was in nutritional indices of *B. lacerta* except for AI. The most significant difference was between winter and summer for AI (Table 5,  $p<0.05$ ).

**Table 4.** Seasonally fatty acid composition for gonads of *B. lacerta* (% of total FA)

FA	GONAD (Ovary)			
	Spring (n=6)	Summer (n=6)	Autumn (n=10)	Winter (n=10)
12:0	1.50±1.20	1.90±1.60	0.77±0.73	0.88±0.53
14:0	3.84±2.68 <sup>ab</sup>	5.67±2.16 <sup>a</sup>	2.49±0.82 <sup>b</sup>	2.37±1.78 <sup>b</sup>
16:0	0.86±0.97 <sup>b</sup>	-	1.04±1.06 <sup>b</sup>	1.99±1.02 <sup>a</sup>
15:0	-	-	0.52±0.30	0.81±0.46
16:0	16.82±4.36	20.28±4.75	17.53±4.16	19.01±4.28
17:0	-	1.39±0.71	0.51±0.32	0.62±0.36
18:0	4.86±2.64	3.87±2.22	4.98±3.72	2.71±3.75
20:0	1.31±0.73	-	0.64±0.48	0.61±0.28
23:0	-	-	-	-
*MSFA	1.17±0.87 <sup>a</sup>	1.25±0.79 <sup>a</sup>	0.44±0.28 <sup>b</sup>	0.62±0.41 <sup>b</sup>
ΣSFA	30.36±1.03	34.36±2.15	28.92±3.12	29.62±0.87
14:1	0.62±0.43	1.09±0.76	0.59±0.32	0.86±0.68
16:1ω7	12.23±1.42	11.82±3.36	14.11±5.67	15.58±4.87
17:1	-	-	-	-
18:1ω9	6.95±3.32	8.41±3.38	13.60±8.25	10.41±3.75
20:1ω9	1.46±0.96	1.23±0.85	1.22±0.71	0.72±0.42
22:1ω9	1.47±1.18	1.43±1.15	2.59±3.12	2.27±3.67
22:1ω11	0.72±0.45	0.56±0.24	-	-
*MMUFA	0.52±0.35	-	-	-
ΣMUFA	23.35±5.06	24.54±6.28	32.11±6.12	29.82±5.76
16:2ω4	2.44±1.34	0.96±0.19	0.93±0.54	0.51±0.45
18:2ω6	1.94±0.89	1.73±0.68	2.74±2.24	1.73±1.05
18:3ω3	3.50±1.49	4.85±2.20	4.77±2.58	2.37±1.03
ARA	7.72±4.28	5.06±3.56	7.05±4.92	9.16±3.90
EPA	16.93±4.06 <sup>ab</sup>	17.44±5.25 <sup>b</sup>	11.56±4.91 <sup>a</sup>	15.20±3.50 <sup>ab</sup>
22:2	1.17±0.36	1.17±0.41	1.13±0.39	1.10±0.66
DHA	11.80±6.71	8.86±4.75	9.79±5.60	9.80±4.34
*MPUFA	1.40±0.81	1.03±0.42	1.00±0.57	0.69±0.36
ΣPUFA	46.29±4.56	41.10±5.83	38.97±3.85	40.56±3.45
Σω3	32.23±7.27	31.15±5.12	26.12±1.39	27.37±2.45
DHA/EPA	0.81±0.22	0.53±0.19	0.85±0.24	0.65±0.27

\*Minor FA<0.5 (19:0, 21:0, 23:0 from SFA; 15:1, 20:1ω7, 20:1ω11 from MUFA; 18:3ω4, 18:3ω6, 20:2ω6, 22:5ω3 from PUFA) ; Means followed by different letters, letter groups in the same row are significantly different ( $p<0.05$ ), while means do not differ if there are no letters in the same row ( $p<0.05$ ). ± SD (Standard Deviation), '-' non defined.

**Table 5.** Seasonally nutritional quality index fatty acids in muscle of *B. lacerta* (% of total FA)

FAs	Spring (n=12)	Summer (n=9)	Autumn (n=14)	Winter (n=18)
AI	0.47±0.18 <sup>ab</sup>	0.57±0.15 <sup>b</sup>	0.42±0.12 <sup>a</sup>	0.41±0.09 <sup>a</sup>
TI	0.54±0.08	0.55±0.11	0.58±0.07	0.56±0.08
h/H	3.10±0.77	2.40±1.07	4.47±2.61	2.73±0.51
OFAs	20.34±3.45	24.34±5.23	19.77±5.59	21.53±2.79
DFA	75.51±14.78	72.10±24.00	77.18±13.91	74.05±17.91
HPI	2.59±0.82	2.04±0.70	3.00±1.56	2.76±0.72
ΣPUFA/ΣSFA	2.05±0.50	1.68±0.68	2.08±1.41	1.90±0.44
EPA+DHA	36.24±4.31	30.03±4.79	28.67±6.04	30.75±6.36
ω6/ω3	0.18±0.11	0.16±0.06	0.19±0.09	0.23±0.09

\*Means followed by different letters (a, b), letter groups (ab) in the same row are significantly different ( $p<0.05$ ), while means do not differ if there are no letters in the same row ( $p<0.05$ ). ± SD (Standard Deviation).

## DISCUSSION

There were differences among the FA composition of the tissues/organs of wild female *B. lacerta*. The most important difference was between muscle and gonad. However, these differences were not valid for all fatty acids. 14:0, 16:0, 16:1 $\omega$ 7, 18:1 $\omega$ 9, DHA, EPA, ARA were in the foreground in the three tissues in PCO analysis in *B. lacerta*. 14:0, 16:1 $\omega$ 7, 18:3 $\omega$ 3, 18:2 $\omega$ 6 (Linoleic acid, LA) and ARA have been reported to be typical freshwater fatty acids (Parzanini et al., 2021). Ugoala et al. (2009) reported that the predominant fatty acids in freshwater fish are 14:0 and 16:0 from SFA and 18:1 from MUFA. The predominant PUFA is from the  $\omega$ 6 series and is mainly 18:2 FA. In addition, EFA compounds 18:3 $\omega$ 3 (Alpha-linolenic Acid, ALA) and 18:2 $\omega$ 6 are importance in PUFA. However, freshwater fish have an irregular FA pattern, which are good sources of  $\omega$ 6 EFA (Ugoala et al., 2009). We showed that *B. lacerta* had higher percentage of  $\omega$ 3 FA than  $\omega$ 6 FA. There were only two major fatty acids, ARA and LA from  $\omega$ 6 FA. A similar result was found by Parzanini et al. (2021) in tissues/organs (muscle, liver and eyes) of the European eel. They showed that  $\omega$ 3 FA were higher than  $\omega$ 6 FA in the tissues/organs in all circumstances. On the other hand, it was found that the balanced ratio of  $\omega$ 3 and  $\omega$ 6 FA in freshwater fish increased the reproductive performance and hatchability of zebrafish, *Danio rerio* females by Jaya-Ram et al. (2008). In this study, the most important differences among the tissues/organs were seen in  $\Sigma$ PUFA and  $\Sigma$  $\omega$ 3 FA and ARA, EPA and DHA from PUFA. Also, the change of 16:0 from SFA between the tissues/organs was important. ARA was the lowest in the liver, the highest in the gonad, while the EPA and DHA were lower in the gonad, and the highest EPA was in the liver, the lowest DHA was in the muscle tissue. However, the percentage of LC-PUFA in the muscle and liver of *B. lacerta* was higher than gonad. The possible reason for the high percentage of this situation in the muscle of fish is that the LC-PUFA serves as the main energy provider in fish muscle (Hong et al., 2014) and liver (Tocher, 2003). Some recent studies highlighted the key role of LC-PUFA, particularly ARA, in regulating physiological functions of reproduction (Tocher et al., 2010; Majdoubi et al., 2020). Gonad fatty acid composition varies within each mature stage (Anido et al., 2015). The spawning period of *B. lacerta* generally occurs from late March to August with a peak in April (Dopeikar et al., 2015). Highest ARA in gonad of *B. lacerta* was in winter (9%) and spring (8%). ARA is the precursor of several eicosanoids which are produced by the ovarian tissues and play an important role in the ovulation process (Suloma and Ogata, 2011). Thus, ARA was higher in spawning seasons than the other seasons (Majdoubi et al., 2020). Stream invertebrates have a limited innate ability to transform ALA to EPA (trophic upgrading). Thus, their PUFA composition mostly resembles dietary PUFA (Masclaux et al., 2012). Probably, the EPA content of *B. lacerta* was related to terrestrial transport as well as the rich benthic fauna of the Murat River, because EPA percentages of three tissues/organs were higher than the other seasons in spring and summer. In particular, primary production increases in the

Murat River due to the excess of terrestrial origin transport in the spring season, which ensures the enrichment of benthic fauna and the fatty acid composition of *B. lacerta*, a benthopelagic freshwater fish, is also affected. 16:1 $\omega$ 7 was the first high abundance MUFA (12-14%) in *B. lacerta* the tissues/organs. The second highest abundance MUFA was 18:1 $\omega$ 9 (9-11%). In contrast, some studies reported that 18:1 $\omega$ 9 was the high abundance MUFA in total lipids in freshwater fish species including the genus *Barbus* (Olgunoğlu et al., 2011; Gokce et al., 2011).

ARA, EPA and DHA proved to be the three most abundant members for *Barbus barbus* (Mancini et al., 2011). Similarly, it was found that EPA, DHA, and ARA were the most abundant fatty acids. Also, 18:1 $\omega$ 9, 16:1 $\omega$ 7 and 16:0 were the most abundant fatty acids in all the three tissues/organs of *B. lacerta* in all seasons. In a study conducted by Gokce et al. (2011) in the lake located at the Euphrates River in South Eastern Türkiye, was reported that MUFA were the highest in the *Barbus crypus* muscle tissue, followed by SFA and PUFA. Similarly, Bayir et al. (2011) found that PUFA were at the lowest percentage in muscle tissue of *Barbus capito capito* in different lipid fractions (polar and neutral lipids) for all seasons in Aras River, Türkiye. In our study, the mean highest PUFA were found in muscle tissue. However, the period in which the study was May and June, which is the breeding season of *Barbus* species (Bayir et al., 2011). If we make a comparison based on the spring period, the most abundant FA group in the spring in the muscle tissue was  $\Sigma$ PUFA (51%),  $\Sigma$ SFA (29%) and  $\Sigma$ MUFA (23%). Although *B. lacerta*, *B. capito capito*, *B. crypus* are members of the same genus (*Barbus*), it is possible that there are some differences due to the location difference and different *Barbus* species.

### Seasonally nutritional indices in muscle of *B. lacerta*

We examined the FA used as food quality indexes such as AI, TI, h/H, OFAs, DFA, HP,  $\Sigma$ PUFA/ $\Sigma$ SFA, EPA+DHA,  $\omega$ 6/ $\omega$ 3 to examine the seasonal changes in nutritional quality of *B. lacerta*. Studies evaluating the nutritional quality of fish based on the food quality index fatty acids are very limited, and no information on this subject has been found for *B. lacerta*. Thus, the evaluation was made by comparing with a limited number of studies conducted with other fish species.

This study determined that there were seasonal changes in the food quality index FA, but the most significant changes were in AI. No difference was found in terms of other indices. The low AI value indicates that the tissues of the examined fish are beneficial for health (Łuczyńska and Paszczyk, 2019). It was determined the lowest AI values in spring (0.42) and winter (0.41) while highest AI value was in summer (0.57). Therefore, we can say that the highest quality *B. lacerta* in terms of AI was found in winter and spring periods. It was reported as 0.37-0.42 in *Abramis brama*, 0.36 in *Cyprinus caprio*, 0.33 in *Oncorhynchus mykiss*, 0.37 in *Perca fluviatilis* (Łuczyńska et al., 2017), 0.64-0.72 in *Salmo trutta* (Dal Bosco et al., 2013), 0.37-0.67 in *Cyprinion macrostomus* (Şen Özdemir et al., 2023)

and 0.29-0.68 in *Micropterus salmoides* (Subhadra et al., 2006) when we looked at the studies conducted with some other freshwater fish. In other indices, seasonal differences within season were not significant and high similarity rates were found in SIMPER analyses and the food quality index that contributed the most to this was fatty acids DFA (55%) and EPA+DHA (22%). The DFA (neutral and hypocholesterolemic fatty acids) index reports the hypocholesterolemic (total cholesterol-lowering) properties of the analyzed lipids (Batkowska et al., 2021). Therefore, the periods when DFA is high represent the most suitable periods for food consumption. Although we did not detect a significant difference among the seasons, we can say that the most suitable consumption periods of *B. lacerta* are autumn (77%) and spring (75%). EPA + DHA is a globally recognized index. The World Health Organization (WHO) and the Food and Agriculture Organization (FAO) recommended an intake of 0.25-2 g EPA + DHA per day (FAO, WHO, 2010). This index is mostly used to evaluate the nutritional value of seafood, especially fish, due to the low content of EPA and DHA in terrestrial plants and animals. Rincón-Cervera et al. (2020) studied the FA composition of fish and shellfish caught in the South Pacific. Their results showed that EPA + DHA varied between 115.15 and 1370.67 mg/100 g in all the studied fish species (Rincón-Cervera et al., 2020). However, there are limited studies on DHA+EPA as a nutritional quality index in inland fish. The EPA+DHA value given as food quality index in fresh water fish *Megalobrama amblycephala* was 5.52-7.36 (Xu et al., 2017). This value was quite low compared to the EPA+DHA of *B. lacerta*. This may indicate that *B. lacerta* has a moderate quality in terms of EPA+DHA as a freshwater fish. According to SIMPER results; EPA+DHA that contributed as the third contributor the difference among the seasons were OFA except for spring and winter. We know that high OFA content reduces the lipid quality of food. In this study, OFA content of *B. lacerta* was relatively high (20-24 %), and spring and autumn were the periods when it was relatively low. TI from the other food quality index FA characterizes the thrombogenic potential of FA, indicating the tendency to form clots in blood vessels and provides the contribution of different FA indicating the relationship between pro-thrombogenic FA (12:0, 14:0 and 16:0) and anti-thrombogenic FA (MUFA and  $\omega$ 3,  $\omega$ 6 families) (Ulbricht et al., 1991). Therefore, consumption of foods or products with lower TI is beneficial for cardiovascular diseases (CVD). In studies on freshwater fish, TI was reported to be 0.16 in *Oncorhynchus mykiss*, 0.20 in *Perca fluviatilis*, 0.18 in *Esox lucius* (Łuczynska et al., 2017), 0.21-0.30 in *Salmo trutta* (Dal Bosco et al., 2013), 0.82-0.87 in *Oreochromis niloticus* (Tonial et al., 2014) and 0.26-0.39 in *Cyprinion macrostomus* (Şen Özdemir et al., 2023). We determined the TI value in *B. lacerta* to be between 0.53-0.58 (winter-autumn). When we compare these values with other reported freshwater fish, it is seen that they are at medium level and low TI increases the nutritional quality of *B. lacerta*. H/h is based on research on the regulation of dietary FA and plasma low density cholesterol (LDL-C) (Dietschy, 1998). It characterizes the relationship between hypocholesterolemic (18:1 and PUFA) and hypercholesterolemic

FA. It is an important nutritional quality index reflecting the effect of FA composition on CVD (Chen and Liu, 2020). h/H changed between 2.40 (summer) and 4.47 (autumn) in *B. lacerta* edible muscle tissue. The h/H value was reported as 1.56-1.63 in freshwater fish *Oreochromis niloticus* (Tonial et al., 2014), 1.88-2.16 in *Salmo trutta* (Dal Bosco et al., 2013) and 1.34-2.20 in *Cyprinion macrostomus* (Şen Özdemir et al., 2023). We saw that *B. lacerta* is a very high-quality food in terms of h/H. Especially, autumn was seen as the best period for consuming *B. lacerta* in terms of h/H.

$\omega$ 6/ $\omega$ 3 is a useful indicator of the nutritional value of fish lipids and a lower ratio is more effective in preventing CVD associated with plasma lipid levels (Rhee et al., 2017).  $\omega$ 6/ $\omega$ 3 should not exceed 5.0 in the human diet. Therefore, it is suggested that increasing  $\omega$ 3 and decreasing  $\omega$ 6 consumptions to decrease the  $\omega$ 6/ $\omega$ 3 ratio is beneficial to human health (Fernandes et al., 2014). Here, it is not the excess of  $\omega$ 6 in the diet but rather the deficiency of  $\omega$ 3 that increases this risk (FAO, 2014). Matos et al. (2019) reported that the  $\omega$ 6/ $\omega$ 3 ratio was 8.16 for Nile tilapia (cage), 5.40 for Common carp (5.40), 5.27 for Grass carp.  $\omega$ 6/ $\omega$ 3 was reported in 0.22-0.29 for *Cyprinion macrostomus* by Şen Özdemir et al. (2023). In the study, the highest  $\omega$ 6/ $\omega$ 3 for *B. lacerta* was determined in winter (0.23) while the lowest  $\omega$ 6/ $\omega$ 3 of *B. lacerta* was in summer (0.16). The values did not exceed 5.0 and  $\Sigma\omega$ 3 fatty acids were higher than  $\Sigma\omega$ 6 FA, since high  $\omega$ 3 and low  $\omega$ 6 increase the nutritional quality of the diet, *B. lacerta* appears to be a quality food source. Additionally, the higher the PUFA/SFA ratio will be the higher positive effect (Liu and Chen, 2020). Foods with a PUFA/SFA ratio below 0.45 are considered undesirable foods for the human diet due to their potential to trigger an increase in cholesterol in the blood (Kromhout 2010). In this study, PUFA/SFA ratio was found to be above 0.45 (1.68-2.08; spring-autumn) in *B. lacerta* for every period.

## CONCLUSIONS

In the study, muscle, gonad and liver tissues were used as factors to determine seasonal changes of *B. lacerta*. Additionally, by determining the food quality index fatty acids (AI, TI, h/H, OFA, DFA, HPI,  $\Sigma$ PUFA/ $\Sigma$ SFA, EPA+DHA,  $\omega$ 6/ $\omega$ 3) in edible muscle tissue, the healthiest consumption period was tried to be determined.

The study indicated that although FA of all the tissue/organs for *B. lacerta* were differed, the most important difference was between muscle and gonads. The seasonal FA composition of each tissue/organ of *B. lacerta* changes in *B. lacerta*. It was observed that ARA was higher in the gonads than the other tissues during the periods when spawning was active, such as the end of winter and the beginning of spring. Because, muscle and liver lipids are mostly used for energy, while gonadal lipids are used for reproduction.  $\omega$ 3 FA were more abundant than  $\omega$ 6 fatty acids. The most abundant  $\omega$ 6 fatty acids were ARA and LA. The most abundant  $\omega$ 3 fatty acids were DHA and EPA.



It was seen that the highest seasonal change was in AI, when the seasonal changes of the food quality index fatty acids were evaluated in edible muscle tissue. We determined that different indexes came to the fore in different seasons except summer, when the food quality index FA were evaluated as a whole. Therefore, it can be suggested that *B. lacerta* can be consumed as a high quality and healthy food partially except summer. It can be seen that *B. lacerta* has a rich PUFA content in all three tissues in all seasons. And this makes it a healthy food that protects against cardiovascular, immune system diseases and high cholesterol.

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### REFERENCES

- Ahmed, I., Jan, K., Fatma, S., & Dawood, M.A. (2022). Muscle proximate composition of various food fish species and their nutritional significance: A review. *Journal of Animal Physiology and Animal Nutrition*, 106(3), 690-719. <https://doi.org/10.1111/jpn.13711>
- Anido, R. V., Zaniboni-Filho, E., Garcia, A.S., Baggio, S.R., & Fracalossi, D.M. (2015). Characterization of the ovary fatty acids composition of *Rhamdia quelen* (Quoy and Gaimard) (Teleostei: Siluriformes), throughout their reproductive cycle. *Neotropical Ichthyology*, 13(2), 453-460. <https://doi.org/10.1590/1982-0224-20140139>
- Batkowska, J., Drabik, K., Brodacki, A., Czech, A., & Adamczuk, A. (2021). Fatty acids profile, cholesterol level and quality of table eggs from hens fed with the addition of linseed and soybean oil. *Food Chemistry*, 334, 127612. <https://doi.org/10.1016/j.foodchem.2020.127612>
- Bayır, A., Sirkecioğlu, A.N., Aksakal, E., Bayır, M., Haliloğlu, H.Y., Güneş, M., & Aras, N.M. (2011). Changes in the fatty acids of neutral and polar lipids of *Silurus glanis* and *Barbus capito capito* during an annual cycle. *Italian Journal of Food Science*, 23(2), 173-179.
- Breslow, J.L. (2006). n-3 Fatty acids and cardiovascular disease. *The American Journal of Clinical Nutrition*, 83(6), 1477S-1482S. <https://doi.org/10.1093/ajcn/83.6.1477S>
- Chen, J., & Liu, H. (2020). Nutritional indices for assessing fatty acids: A mini review. *International Journal of Molecular Sciences*, 21(16), 5695. <https://doi.org/10.3390/ijms21165695>
- Costa, R.G., Mesquita, Í.V.U., Queiroga, R.D.C.R.D.E., Medeiros, A.N.D., Carvalho, F.F.R.D., & Beltrão Filho, E. M. (2008). Características químicas e sensoriais do leite de cabras Moxotó alimentadas com silagem de maniçoba. *Revista Brasileira de Zootecnia*, 37, 694-702. <https://doi.org/10.1590/S1516-35982008000400016>
- Dal Bosco, A.D., Mugnai, C., Roscini, V., & Castellini, C. (2013). Fillet fatty acid composition, estimated indexes of lipid metabolism and oxidative status of wild and farmed brown trout (*Salmo trutta* L.). *Italian Journal of Food Science/Rivista Italiana di Scienza degli Alimenti*, 25(1), 83-89.
- David, F., Sandra, P., & Vickers, A. K. (2005). Column selection for the analysis of fatty acid methyl esters. *Food Analysis Application*. Palo Alto, CA: Agilent Technologies, 19, 19.
- Dopeikar, H., Keivany, Y., & Shadkhast, M. (2015). Reproductive biology and gonad histology of the kura barbel, *Barbus lacerta* (Cyprinidae), in Bibi-Sayyedana River, Tigris basin, Iran. *North-Western Journal of Zoology*, 11, 163-170.
- Dietschy, J.M. (1998). Dietary fatty acids and the regulation of plasma low

### AUTHORSHIP CONTRIBUTIONS

Nurgül Şen Özdemir and Mustafa Koyun contributed to the design and implementation of the study. Also, Mustafa Koyun bought fish samples from fisherman from the sampling area and prepared the catching fish for analysis. Nurgül Şen Özdemir was responsible for data curation and analysis, and writing the original draft of the manuscript. All the authors reviewed and edited the draft.

### CONFLICT OF INTEREST

The authors declare no competing interests.

### ETHICS APPROVAL

No ethical approval was required, since fish samples were obtained from commercial.

### DATA AVAILABILITY

No datasets were generated or analyzed during the current study.

density lipoprotein cholesterol concentrations. *The Journal of Nutrition*, 128(2), 444S-448S. <https://doi.org/10.1093/jn/128.2.444S>

FAO. (2014). The state of world fisheries and aquaculture. Opportunities and challenges. *Food and agriculture Organization of the United Nations*. Rome, Italy, 243 p.

FAO, WHO. (2010). Fats and Fatty Acids in Human Nutrition: Report of an Expert Consultation; FAO food and nutrition paper #91; FAO: Rome, Italy; WHO: Geneva, Switzerland, 21812367.

Fernandes, C.E., da Silva Vasconcelos, M.A., de Almeida Ribeiro, M., Sarubbo, L.A., Andrade, S.A.C., & de Melo Filho, A.B. (2014). Nutritional and lipid profiles in marine fish species from Brazil. *Food Chemistry*, 160, 67-71. <http://dx.doi.org/10.1016/j.foodchem.2014.03.055>

Galloway, A.W., & Winder, M. (2015). Partitioning the relative importance of phylogeny and environmental conditions on phytoplankton fatty acids. *PLoS One*, 10(6), e0130053. <https://doi.org/10.1371/journal.pone.0130053>

Garaffo, M.A., Vassallo-Agius, R., Nengas, Y., Lembo, E., Rando, R., Maisano, R., Dugo, G., & Giuffrida, D. (2011). Fatty acids profile, atherogenic (IA) and thrombogenic (IT) health lipid indices, of raw roe of blue fin tuna (*Thunnus thynnus* L.) and their salted product "Bottarga". *Food and Nutrition Sciences*, 2(7), 736-743. <https://doi.org/10.4236/fns.2011.27101>

Geldiay, R., Balık, S. (2007). *Freshwater fishes of Türkiye*. Ege University Faculty of Fisheries Publications, No: 46. Ege University Press. Bornova İzmir; p.532. (in Turkish)

Gladyshev, M.I., Sushchik, N.N., & Makhutova, O.N. (2013). Production of EPA and DHA in aquatic ecosystems and their transfer to the land. *Prostaglandins & Other Lipid Mediators*, 107, 117-126. <https://doi.org/10.1016/j.prostaglandins.2013.03.002>

Gokce, M.A., Tasbozan, O., Surhan, S.T., Celik, M., Ozcan, F., & Basusta, A. (2011). Proximate composition and fatty acid profile of shabbout (*Barbus grypus* Heckel) caught from the Atatürk Dam Lake, Turkey. *Journal of Food, Agriculture & Environment*, 9(2), 148-151.

Hansen, H.J., & Abraham, S. (1983) Influence of temperature, environmental salinity and fasting on the patterns of fatty acids synthesized by gills and liver of the European eel (*Anguilla Anguilla*). *Comparative Biochemistry and Physiology Part B: Comparative Biochemistry*, 75(4), 581-587. [https://doi.org/10.1016/0305-0491\(83\)90100-1](https://doi.org/10.1016/0305-0491(83)90100-1)

Hara, A., & Radin N.S. (1978) Lipid extraction of tissues with a low-toxicity solvent. *Analytical Biochemistry*, 90(1), 420-426. [https://doi.org/10.1016/0003-2697\(78\)90046-5](https://doi.org/10.1016/0003-2697(78)90046-5)

- Hong, H., Zhou, Y., Wu, H., Luo, Y., & Shen, H. (2014). Lipid content and fatty acid profile of muscle, brain and eyes of seven freshwater fish: A comparative study. *Journal of the American Oil Chemists' Society*, 91, 795-804. <https://doi.org/10.1007/s11746-014-2414-5>
- Jardine, T.D., Galloway, A.W.E., & Kainz, M.J. (2020). Unlocking the power of fatty acids as dietary tracers and metabolic signals in fishes and aquatic invertebrates. *Philosophical Transactions of the Royal Society B-Biological Sciences*, 375, 11. <https://doi.org/10.1098/rstb.2019.0639>
- Jaya-Ram, A., Kuah, M.K., Lim, P.S., Kolkovski, S., & Shu-Chien, A.C. (2008). Influence of dietary HUFA levels on reproductive performance, tissue fatty acid profile and desaturase and elongase mRNAs expression in female zebrafish *Danio rerio*. *Aquaculture*, 277(3-4), 275-281. <http://dx.doi.org/10.1093/cdn/nzaa034>
- Kromhout, D., Giltay, E.J., & Geleijnse, J.M. (2010). n-3 Fatty acids and cardiovascular events after myocardial infarction. *New England Journal of Medicine*, 363(21), 2015-2026. <http://dx.doi.org/10.1056/NEJMoa1003603>
- Liu, W., Lyu, J., Wu, D., Cao, Y., Ma, Q., Lu, Y., & Zhang, X. (2022). Cutting techniques in the fish industry: A critical review. *Foods*, 11(20), 3206. <https://doi.org/10.3390/foods11203206>
- Łuczyńska, J., & Paszczyk, B. (2019). Health risk assessment of heavy metals and lipid quality indexes in freshwater fish from lakes of Warmia and Mazury region, Poland. *International Journal of Environmental Research and Public Health*, 16(19), 3780. <https://doi.org/10.3390/ijerph16193780>
- Łuczyńska, J., Paszczyk, B., Nowosad, J., & Łuczyński, M.J. (2017). Mercury, fatty acids content and lipid quality indexes in muscles of freshwater and marine fish on the polish market. Risk assessment of fish consumption. *International Journal of Environmental Research and Public Health*, 14(10), 1120. <https://doi.org/10.3390/ijerph14101120>
- Majdoubi, F.Z., Benhima, R., Ouizgane, A., Farid, S., Droussi, M., Gueriero, G., & Hasnaoui, M. (2020). Ova Fatty Acids Composition and Spawning Performances of Silver Carp, *Hypophthalmichthys molitrix* (Morocco). *Turkish Journal of Aquatic Sciences*, 20(12), 879-888. [https://doi.org/10.4194/1303-2712-v20\\_12\\_04](https://doi.org/10.4194/1303-2712-v20_12_04)
- Mancini, I., Defant, A., Mesarić, T., Potočnik, F., Batista, U., Guella, G., ... & Sepčić, K. (2011). Fatty acid composition of common barbel (*Barbus barbus*) roe and evaluation of its haemolytic and cytotoxic activities. *Toxicon*, 57(7-8), 1017-1022. <https://doi.org/10.1016/j.toxicon.2011.04.004>
- Masclaux, H., Bec, A., Kainz, M.J., Perriere, F., Desvillettes, C., & Bourdier, G. (2012). Accumulation of polyunsaturated fatty acids by cladocerans: effects of taxonomy, temperature and food. *Freshwater Biology*, 57, 696-703. <https://doi.org/10.1111/j.1365-2427.2012.02735.x>
- Matos, A. P., Matos, A. C., & Moecke, E. H. S. (2019). Polyunsaturated fatty acids and nutritional quality of five freshwater fish species cultivated in the western region of Santa Catarina, Brazil. *Brazilian Journal of Food Technology*, 22, e2018193. <https://doi.org/10.1590/1981-6723.19318>
- Mițitelu, M., Lupuliasa, D., Neacșu, S. M., Olteanu, G., Busnatu, Ș. S., Mihai, A., Popovici, V., Măru, N., Boroghina, S.C., Mihai, S., Ioniță-Mîndrican, C.B., & Scafa-Udrishte, A. (2024). Polyunsaturated fatty acids and human health: A key to modern nutritional balance in association with polyphenolic compounds from food sources. *Foods*, 14(1), 46. <https://doi.org/10.3390/foods14010046>
- Olgunoğlu, I. A., Olgunoğlu, M. P., & Artar, E. (2011). Seasonally changes in biochemical composition and meat yield of shabut (*Barbus grypus*, Heckel 1843. *Iranian Journal of Fisheries Sciences*, 10(1), 181-187.
- Parzanini, C., Arts, M. T., Rohla, M., Koprivnikar, J., Power, M., Skiftesvik, A. B., Browman, H. I., Milotic, D., & Durif, C. M. F. (2021). Feeding habitat and silencing stage affect lipid content and fatty acid composition of European eel *Anguilla anguilla* tissues. *Journal of Fish Biology*, 99, 1110-1124. <https://doi.org/10.1111/jfb.14815>
- Pierron, F., Baudrimont, M., Dufour, S., Elie, P., Bossy, A., Lucia, M., & Massabuau, J.C. (2009). Ovarian gene transcription and effect of cadmium pre-exposure during artificial sexual maturation of the European eel (*Anguilla anguilla*). *Biomaterials*, 22, 985-994. <http://dx.doi.org/10.1007/s10534-009-9250-3>
- Rhee, J.J., Kim, E., Buring, J.E., & Kurth, T. (2017). Fish consumption, omega-3 fatty acids, and risk of cardiovascular disease. *American Journal of Preventive Medicine*, 52(1), 10-19. <http://dx.doi.org/10.1016/j.amepre.2016.07.020>
- Rincón-Cervera, M.Á., González-Barriga, V., Romero, J., Rojas, R., & López-Arana, S. (2020). Quantification and distribution of omega-3 fatty acids in South Pacific fish and shellfish species. *Foods*, 9(2), 233. <https://doi.org/10.3390/foods9020233>
- Santos-Silva, J., Bessa, R.J.B., & Santos-Silva, F.J.L.P.S. (2002). Effect of genotype, feeding system and slaughter weight on the quality of light lambs: II. Fatty acid composition of meat. *Livestock Production Science*, 77(2-3), 187-194. [https://doi.org/10.1016/S03016226\(02\)00059-3](https://doi.org/10.1016/S03016226(02)00059-3)
- Sargent, J. R., Tocher, D.R., & Bell, J.G., 2002. The lipids. In: Halver, J. E. & Hardy, R.W. (Eds.), *Fish nutrition*, pp. 181- 257, 3rd San Diego, Academic Press.
- Silva, A.T., Ruvieri, E.S.P., Mateus O., Magalhaes, M.O., Neto, N.L., & Houngru, K. (2019). An alise Fatorial Exploratória: aplica ç ao em dados de crismescontra as mulheres em 141 munic ípios do estado de Mato Grosso no ano de 2016. *Alfenas*, 8(2), 427-436 (in Spanish).
- Suloma, A., & Ogata, H.Y. (2011). Arachidonic acid is a major component in gonadal fatty acids of tropical coral reef fish in the Philippines and Japan. *Journal of Aquatic Research & Development*, 2, 111. <https://doi.org/10.4172/2155-9546.1000111>
- Subhadra, B., Lochmann, R., Rawles, S., & Chen, R. (2006). Effect of dietary lipid source on the growth, tissue composition and hematological parameters of largemouth bass (*Micropterus salmoides*). *Aquaculture*, 255(1-4), 210-222. <https://doi.org/10.1016/j.aquaculture.2005.11.043>
- Şen Özdemir, N.Ş., Koyun, M., Caf, F., & Kırıcı, M. (2023). Factors affecting nutritional quality in terms of the fatty acid composition of *Cyprinion macrostomus*. *Grasas y Aceites*, 74(2), e508-e508. <https://doi.org/10.3989/gya.0444221>
- Tibaoui, S., Smeti, S., Essid, I., Bertolin, J.R., Joy, M., & Atti, N. (2020). Physicochemical characteristics, fatty acid profile, alpha-tocopherol content, and lipid oxidation of meat from ewes fed different levels of distilled myrtle residues. *Molecules*, 25(21), 4975. <https://doi.org/10.3390/molecules25214975>
- Tocher, D.R. (2003). Metabolism and functions of lipids and fatty acids in teleost fish. *Reviews in Fisheries Science*, 11(2), 107-184. <https://doi.org/10.1080/713610925>
- Tocher, D.R. (2010). Fatty acid requirements in ontogeny of marine and freshwater fish. *Aquaculture Research*, 41, 717-732. <https://doi.org/10.1111/j.1365-2109.2008.02150.x>
- Tonial, I.B., Oliveira, D.D., Coelho, A.R., Matsushita, M., Coró, F.A.G., Souza, N.D., & Visentainer, J.V., 2014. Quantification of essential fatty acids and assessment of the nutritional quality indexes of lipids in tilapia alevins and juvenile tilapia fish (*Oreochromis niloticus*). *Journal of Food Research*, 3(3), 105-114. <https://doi.org/10.5539/jfr.v3n3p105>
- Ugoala, C., Ndukwe, G.I. & Audu, T.O. (2009). Fatty acids composition and nutritional quality of some freshwater fishes. *Nature Precedings*, <https://doi.org/10.1038/npre.2009.3239.1>
- Ulbricht, T.L.V., & Southgate, D.A.T. (1991). Coronary heart disease: seven dietary factors. *The Lancet*, 338(8773), 985-992. [https://doi.org/10.1016/0140-6736\(91\)91846-M](https://doi.org/10.1016/0140-6736(91)91846-M)
- Xu WeiNa, X.W., Qian Yu, Q.Y., Li XiangFei, L.X., Li JunYi, L.J., Li PengFei, L.P., Cai DongSen, C.D., & Liu WenBin, L.W. (2017). Effects of dietary biotin on growth performance and fatty acids metabolism in blunt snout bream, *Megalobrama amblycephala* fed with different lipid levels diets. *Aquaculture*, 479, 790-797. <https://doi.org/10.1016/j.aquaculture.2017.07.018>