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RESEARCH ARTICLE

Chemical Composition of Two Grey Mullet Species (*Chelon auratus*, *Mugil cephalus*): A Comparative Study on Wild and Aquaculture-Adapted Species

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Abstract: This study assessed the chemical composition of wild and culture-adapted golden grey mullet (*Chelon auratus*) and flathead grey mullet (*Mugil cephalus*). Wild samples were collected seasonally from Trabzon shores and compared to those adapted to aquaculture conditions for one year. Both species had high levels of glutamate, aspartate, alanine, leucine, and isoleucine. *Mugil cephalus* was a high protein source for adults, with a digestible indispensable amino acid score (DIAAS) score above 100 in both wild and culture forms. However, a DIAAS score exceeding 100 was only detected in wild *Chelon auratus* in the winter and spring. Both species had high levels of EPA and DHA, with *Mugil cephalus* having the highest lipid quality. Despite seasonal variations, cultured fish maintained a good nutritional profile similar to wild fish, especially in the autumn and summer.

Anahtar kelimeler:

Protein kalitesi
DIAAS
Lipit indeksleri
Karadeniz
Su ürünleri

İki Kefal Türünün (*Chelon auratus*, *Mugil cephalus*) Kimyasal Kompozisyonu: Doğal ve Akuakültüre Adapte Edilmiş Türlerin Karşılaştırılması

Öz: Bu çalışmada, doğal ve kültüre adapte edilmiş altınbaş kefal (*Chelon auratus*) ve has kefal (*Mugil cephalus*) türlerinin kimyasal bileşimi değerlendirilmiştir. Örnekler Trabzon kıyılarından mevsimsel olarak avlanmış ve bir yıl boyunca yetiştiricilik şartlarına adapte edilen bireylerle karşılaştırılmıştır. Her iki türde de yüksek düzeyde glutamat, aspartat, alanin, lösin ve izölösin tespit edilmiştir. *Mugil cephalus*, doğal ve kültür formlarında 100'ün üzerinde bir sindirilebilir elzem amino asit skoruna (DIAAS) sahip yüksek protein kaynağı olarak belirlenmiştir. Ancak, 100'ü aşan DIAAS skoru sadece kış ve ilkbaharda doğadan avlanan *Chelon auratus*'ta tespit edilmiştir. Her iki türde de yüksek miktarda EPA ve DHA tespit edilmiş olup *Mugil cephalus* türünün en yüksek lipit kalitesine sahip olduğu belirlenmiştir. Mevsimsel değişikliklere rağmen, kültüre adapte edilen balıkların, özellikle sonbahar ve yaz aylarında, doğadan avlanan balıklara benzer iyi bir besin profilini koruduğu tespit edilmiştir.

Introduction

The increasing global population necessitates the efficient use of diminishing natural resources, with seafood consistently playing a vital role in human nutrition throughout history. However, overfishing and adverse environmental factors have caused a rapid decline in natural fish populations, with some species even facing extinction. Consequently, aquaculture has become essential to meet the growing demand for food and protein (Lee and Tamaru, 1988; Harmantepe and Büyükhatipoğlu, 2007; FAO, 2020). With the global population currently at 7 billion and projected to reach 8 billion within the next 20 years, the demand for aquaculture products is expected to rise

significantly (FAO, 2020). The surge in aquaculture production has also heightened the demand for fish meal and fish oil, which are critical components in fish feed production. These components, serving as primary sources of protein and fat, have seen significant price increases, substantially raising feed costs (Harmantepe and Büyükhatipoğlu, 2007). The profitability of fish farming is closely linked to the growth rate of the fish; hence, producers aim to bring fish to market quickly to maximize returns (Yiğit and Aral, 1999). As a result, fish species that adapt well to environmental conditions and exhibit rapid growth are preferred for cultivation.

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In this context, it is crucial to investigate species that can diversify the aquaculture sector and thrive under current environmental conditions with minimal fish meal and oil requirements in their feed (Thomas et al., 2021; van Riel et al., 2023). As herbivorous fish, grey mullets are among the promising candidates for such diversification (Lee and Tamaru, 1988) due to their unique tolerance to a wide range of salinity and temperature levels. As pelagic species, grey mullets predominantly inhabit coastal areas of tropical and subtropical seas (Whitfield and Durand, 2023). *Mugil cephalus*, a notable species in this family, is widely distributed in the Black Sea, Mediterranean Sea, Aegean Sea, and along the Spanish coast of the Atlantic Ocean. *Chelon auratus* (Risso, 1810) is found in the Black Sea, the Mediterranean, the Sea of Azov, the southern coasts of Africa and Europe, the Caspian Sea, the Scandinavian coasts and the southern coasts of England. They spend the winter in the warmer waters of Crimea, the Caucasus, and Anatolia in the Black Sea and the Sea of Marmara, migrating northwest and northeast to feed (Çiloğlu, 2023; Froese and Pauly, 2024).

Despite their potential, grey mullet species are not extensively cultured, with only a few scientific research conducted (Garcia-Marquez et al., 2011; Nguyen et al., 2023; Quiros-Pozo et al., 2023). From 2015 to 2019, the TAGEM project "Determination of the Bioecology and Aquaculture Characteristics of the Mullet Species (*Mugil cephalus*, *Liza aurata*) in the Eastern Black Sea" focused on

adapting grey mullet species to culture conditions as well as fish feed (Altuntaş et al., 2020). In the context of this project, grey mullet species were successfully adapted to culture conditions over approximately one year. This study uses both wild and aquaculture-adapted individuals from this project. The main aim of this study is to evaluate the chemical composition of wild and culture-adapted golden grey mullet (*Chelon auratus*) and flathead grey mullet (*Mugil cephalus*) individuals to determine their suitability for aquaculture and to compare the nutritional profiles of cultured and wild individuals across different seasons. Preliminary results indicate that both mullet species have successfully adapted to culture conditions and show significant potential for further rearing activities.

Material and Methods

Fish material and fishing operation

This study examined two economic grey mullet species, golden grey mullet *Chelon auratus* (Risso, 1810) and flathead grey mullet *Mugil cephalus* (Linnaeus, 1758). The study materials were obtained from the Project titled "Determination of the Bioecology and Aquaculture Characteristics of the Mullet Species (*Mugil cephalus*, *Liza aurata*) in the Eastern Black Sea". In the project context, grey mullet species were caught seasonally, and some individuals were adapted to the culture conditions. The workflow of the study is shown in Figure 1.

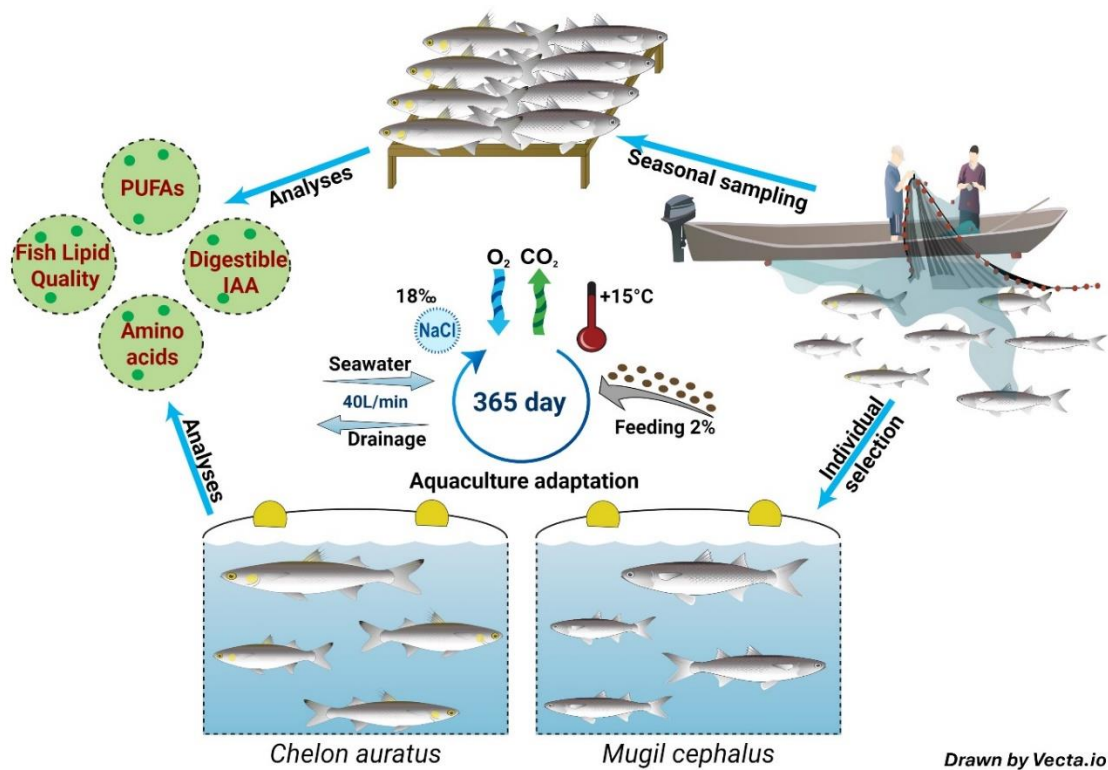


Figure 1. Workflow diagram of this study's methodological procedure. The graphic was drawn using Vecta.io software (Vecta, 2024)

Fishing operations were carried out by partnering with a small-scale fishing boat owner who operated in Trabzon, Eastern Black Sea. Legal permits were provided for the fishing ban period. Surveys were carried out periodically in mids of October, February, April, July from 2016 to 2017 along Trabzon coast. Sampling coordinates were between 41°0'43.79"N, 39°43'30.10"E and 40°58'31.64"N, 39°50'13.24"E. Sampling stations were shown in Figure 2. Fish were caught using gillnet nets with 36 mm mesh size. In sampling studies, two grey mullet species were seen in exact locations every season. Some of the samples were

stored for the chemical analyses at +4 °C, while some were collected as live material for adaptation studies. For aquaculture adaptation studies, the fish's body form and condition were considered, along with their health status after the fishing operation. Individuals who lost scales or were damaged were often lost during adaptation due to infections caused by these injuries. Therefore, only individuals with no signs of external injuries were directly transferred from the ship's deck to special transport tanks (0.5 m³).

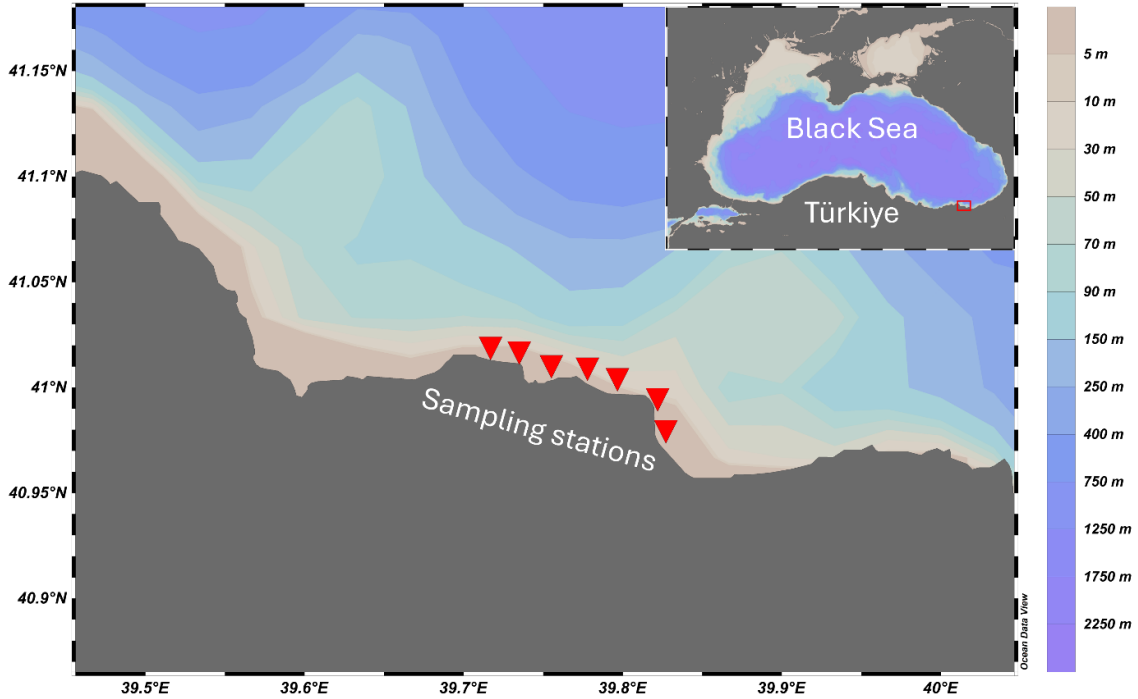


Figure 2. Sampling stations on Trabzon coast of the Black Sea. The sampling map was drawn by ODV software (Schlitzer, 2024)

Aquaculture adaptation and rearing conditions

In this study, a group of fish were selected for aquaculture adaptation studies. Some of them were stored for chemical analysis and immediately transported to the laboratory in cold storage at +4 °C. The fish taken alive from the fishing vehicles were placed in transportation tanks with liquid oxygen and transferred to the marine fish adaptation unit at Central Fisheries Research Institute. Grey mullets were placed in separate fiberglass tanks (2x2x1m) in an open system aquaculture unit with continuous water exchange at 40 L/min. The tanks were supplied with seawater drawn from a depth of 54 m, approximately 1 km off the coast of Trabzon-Yomra. The mean salinity was 18‰ and the temperature was 15 °C. The fish were not fed for the first two days to allow them to adapt to the tank environment. A mixture of feed consisting of small crustaceans, macroalgae, and pelagic fish was provided to adapt them to feeding. From the very first feeding, a meticulous approach was taken. Fresh feed was used for ten days, followed by a commercial pellet fish feed. The fish were fed approximately 2% of their total weight daily, ensuring a consistent and precise feeding regime.

Harvesting and pre-treatment for analyses

After the one-year adaptation study, a subset of the fish was harvested for this study. It is important to note that only fish successfully adapted to the feed and aquaculture environment were used. Throughout the adaptation and harvesting procedures, strict adherence to ARRIVE guidelines (Kilkenny et al., 2010) and EU directive for animal experiments (European Commission, 2010) was maintained, ensuring the highest ethical standards. After harvesting, the fish were anesthetized in clove oil baths (50 µL/L) and euthanized immediately by the gill-cutting method. All wild and aquaculture samples were skinned and filleted, and their internal organs were removed. Finally, the obtained fillets were homogenized at 1800 rpm and stored until the analyses were at +4 °C. In this study, 30 individuals were used in each season and cultured form for each species (n=150 for *C. auratus*, n=150 for *M. cephalus*). Mean length and weight were determined as 28.19±1.14 cm and 367.12±14.06 g for *C. auratus*, and 26.86±0.97 cm and 455.38±11.29 g for *M. cephalus*.

Proximate composition analyses

Moisture analyses were performed using Horwitz's (2000) method. Two grams of the homogenates were placed in petri plates and dried in an oven (Sanyo MOV1125, Japan) at 100 °C for 24 hours. The dried samples were weighed with analytical balance (Kern ABJ-220, Philippines), and the % moisture (water) content was determined. Crude protein analysis was performed using the AOAC (2000) method. 0.5 grams of the samples were weighed, taken into distillation tubes, and burned with 15 mL H₂SO₄ and Kjeldahl tablet at 450 °C. The solutions were distilled with distillation equipment (FOSS 2100, Denmark) and the, obtained solutions were then titrated with 0.1 N HCl, and the protein ratio was calculated. Crude fat analysis was performed according to Folch et al. (1957)'s method. In this method, 0.5-gram samples were weighed, and 10 mL of methanol-chloroform (2:1) mixture was added and kept in the dark for 12 hours. The filtered samples were then concentrated in a Rotary evaporator (Eyela N-N 1521, Japan) to calculate the % of the crude fat content. Crude ash content was determined according to Horwitz's (2000) method. The samples weighed 1 gram in heat-resistant porcelain crucibles and were incinerated in a muffle furnace (Protherm PLF, Türkiye) at 600 °C for 6 hours. The obtained samples were weighed, and crude ash content was determined by percentage.

Amino acid analysis

Acidic digestion was used to prepare the fish meat for analysis. Fish meat was hydrolyzed with HCl until completely hydrolyzed (24 hours, 110 °C) (Çankırılıgil et al., 2020). The hydrolysates obtained were filtered through a 0.45 µm syringe filter and diluted 10⁻¹ with distilled water. The filtrates obtained were then stored in 1.5 mL amber vials until the analyses started. Amino acid analyses were performed according to Henderson et al. (2000). The solutions obtained were filtered through a PTFE injector filter. The filtrates were transferred to 1.5 mL vials and analyzed on an HPLC with a DAD detector (Agilent Infinity II, U.S.A) at 40 °C with a 2 mL/min flow rate. For the separation of amino acids, Zorbax Eclipse AAA amino acid column was used as stationary phase and a mixture (45%:45%:10%) of 40 mM Na₂HPO₄(A) and MeOH:ACN:H₂O(B) fixed at pH 7.8 with 10 N NaOH as mobile phase. The gradient stages of the mobile phase were A:100%, B:0% at the 1.9th minute; A:43%, B:57% at the 18.1st minute; A:0%, B:100% at the 18.6th minute; A:0%, B:100% at the 22.3rd minute and A:100%, B:0% at the 23.2nd minute. Amino acids were detected at 338 nm and 262 nm.

Protein quality evaluation

The protein qualities of both grey mullet species were evaluated using the digestible indispensable amino acid score (DIAAS) method (FAO, 2013). In this method, IAA reference ratios were calculated by multiplying the true ileal digestibility (df) for each indispensable amino acid in the grams of protein in the samples comparing to the reference protein values (FAO, 2013; Kendler et al., 2023). True ileal digestibility reflects the actual amount of amino acid the

metabolism can use. Limited digestibility data exists for fish species individually, including grey-mullet species. This study used universal values for amino acid digestibility of fish muscle described by Moughan et al. (2012). Actual ileal digestibility factors were accepted as 85% for histidine, 93% for isoleucine, 91% for leucine, 93% for lysine, 83% for phenylalanine, 95% for threonine, and 90% for valine. According to the literature, no specific digestibility data exists for cysteine, methionine, and tyrosine in fish muscle. Thus, the true ileal protein digestibility of fish protein used for those amino acids is 90% (Moughan et al., 2012; Shaheen et al., 2016). Reference protein values were obtained from WHO/FAO/UNU (2007) for two distinct age groups: children (6 months to 3 years) and older children, adolescents, and adults. Finally, the lowest DIAA value was multiplied by 100, and the DIAA score (DIAAS) was calculated (FAO, 2013). The equations used in the study are shown below.

$$\text{mg amino acid per g protein} = \frac{\text{mg amino acid per g wet weight}}{\text{g protein per g wet weight}}$$

$$\text{Digestible IAA reference ratio} = \frac{\text{mg of amino acid in 1 g sample protein} \times (\text{df}/100)}{\text{mg of amino acid in 1 g of reference protein}}$$

$$\text{DIAAS} = 100 \times \text{lowest digestible IAA reference ratio of given amino acids}$$

Fatty acid analysis

Fatty acid analysis was performed according to IUPAC (1987). A 0.15 g of the lipid sample was weighed, obtained from the crude fat extraction procedure performed according to Folch et al. (1957). 5 mL of methanolic 0.5 N NaOH and the boiling stone were added into the samples which were then placed in Soxhlet evaporator (Velp Scientifica Ser148, Italy). The temperature was fixed at 65 °C. After 15 minutes, 5 mL BF₃ was added, and the samples were kept at 65 °C for another 2 minutes. At the end of 2 minutes, 2 mL heptane was added to the samples. After 1 minute of evaporation at 65 °C, the obtained mixture was transferred into 25 mL flasks, and they were mixed with saturated NaCl solution to form a liquid phase containing lipids. The obtained phase was filtered with a 0.45 µm injector filter and stored in 1.5 mL vials. Finally, the samples were analyzed in gas chromatography (Shimadzu GC-17A, Japan) with an FID detector and 150 cm column.

Calculation of lipid quality indices

Obtained fatty acid data was evaluated to assess the lipid quality of the grey-mullet species. Total eicosapentaenoic acid and docosahexaenoic content as mg per 100g (EPA+DHA mg/100g), polyunsaturated fatty acid/saturated fatty acid ratio (PUFA/SFA), total omega 3 and omega 6 fatty acid ratio (n-3/n-6), linoleic acid/ α -linolenic acid ratio (LA/ALA), fish lipid quality (FLQ), health-promoting index (HPI), unsaturation index (UI) were calculated according to Chen and Liu (2020). While the nutritional value index (NVI) was calculated using the method described by Wołoszyn et al. (2020), the hypo/hypercholesterolemic ratio (h/H) was determined according to Santos-Silva et al. (2002). Finally, the atherogenicity index (AI) and thrombogenicity index (TI) were calculated according to Ulbricht and Southgate (1991). Equations that were used in assessing lipid quality are shown below:

$$PUFA/SFA = \sum PUFA / \sum SFA$$

$$LA/ALA = C18:2 n - 6 / C18:3 n - 3$$

$$n - 3/n - 6 = \sum n - 3 / \sum n - 6$$

$$NVI = (C18:0 + C18:1) / C16:0$$

$$FLQ = 100 \times (C22:6 n - 3 + C20:5 n - 3) / \sum FA$$

$$HPI = \sum UFA / [C12:0 + (4 \times C14:0) + C16:0]$$

$$UI = 1 \times (\% \text{monoenoics}) + 2 \times (\% \text{dienoics}) + 3 \times (\% \text{trienoics}) + 4 \times (\% \text{tetraenoics}) + 5 \times (\% \text{pentaenoics}) + 6 \times (\% \text{hexaenoics})$$

$$HH = (C18:1 + \sum PUFA) / (C14:0 + C16:0)$$

$$AI = [C12:0 + (4 \times C14:0) + C16:0] / (\sum MUFA + \sum PUFA)$$

$$TI = (C14:0 + C16:0 + C18:0) / [(0.5 \times \sum MUFA) + (0.5 \times \sum n - 6 PUFA) + (3 \times n - 3 PUFA) + (n - 3 / n - 6)]$$

Statistical analysis

Data were expressed as mean values \pm standard error (SE). Differences between mean values of aquaculture forms of the two species were determined via T-test. In contrast, differences between mean values of seasonal variations of the same fish species were evaluated by one-way variance analysis (ANOVA). Homogeneity and normal distribution of the values were calculated using Levene and Anderson-Darling tests, respectively. The significance level (p value) was adopted as 0.05, and mean values less than

0.05 were accepted as statistically significant. The IBM SPSS 23 was used in all statistical analyses.

Results

The proximate composition of grey mullet species was presented in Table 1. It was determined that the moisture (water) was 71.69 \pm 0.27-78.31 \pm 0.38%, crude protein was 16.71 \pm 0.12%-18.62 \pm 0.24%, crude fat was 2.11 \pm 0.18-7.27 \pm 0.27% and crude ash was 1.37 \pm 0.07-1.90 \pm 0.23% in the muscle tissue of golden grey mullet (*Chelon auratus*). The highest crude protein, crude fat and crude ash were determined in spring and winter, respectively. The lowest protein content was found in autumn samples, while the lowest crude fat and crude ash content were found in summer and culture forms (p < 0.05). In muscle tissue of flathead grey mullet (*Mugil cephalus*), the moisture (water) was 73.14 \pm 0.26-77.31 \pm 0.22%, crude protein was 16.24 \pm 0.17-16.81 \pm 0.17%, crude fat was 3.85 \pm 0.08-7.70 \pm 0.26% and crude ash was 1.34 \pm 0.11-1.40 \pm 0.17%. The highest crude protein content was found in the autumn, and the highest crude fat content was found in the winter and spring. The lowest crude protein and fat were detected in the summer samples, and the individuals adapted to the culture conditions (p < 0.05). There was no statistical difference between the samples in crude ash contents (p > 0.05). The proximate composition of culture-adapted grey mullet individuals was statistically similar to those caught in nature in the summer.

Table 1. Proximate composition of wild and culture forms of grey mullet species (%)

Golden grey mullet (<i>Chelon auratus</i>)					
	Autumn	Winter	Spring	Summer	Aquaculture
Moisture	77.91 \pm 0.24 ^{ab}	71.69 \pm 0.27 ^c	71.93 \pm 0.48 ^c	78.31 \pm 0.38 ^a	77.41 \pm 0.20 ^b
Protein	16.71 \pm 0.12 ^c	17.61 \pm 0.19 ^b	18.62 \pm 0.24 ^a	17.13 \pm 0.29 ^{ab}	17.06 \pm 0.15 ^{ab}
Fat	2.76 \pm 0.16 ^c	7.27 \pm 0.27 ^a	6.36 \pm 0.24 ^b	2.11 \pm 0.18 ^d	2.23 \pm 0.21 ^d
Ash	1.52 \pm 0.09 ^b	1.90 \pm 0.23 ^a	1.89 \pm 0.09 ^a	1.37 \pm 0.07 ^c	1.42 \pm 0.18 ^c
Flathead grey mullet (<i>Mugil cephalus</i>)					
	Autumn	Winter	Spring	Summer	Aquaculture
Moisture	75.28 \pm 0.28 ^b	73.18 \pm 0.17 ^c	73.14 \pm 0.26 ^c	77.31 \pm 0.22 ^a	77.20 \pm 0.18 ^a
Protein	16.81 \pm 0.17 ^a	16.51 \pm 0.24 ^{ab}	16.50 \pm 0.14 ^{ab}	16.24 \pm 0.17 ^b	16.30 \pm 0.20 ^b
Fat	5.17 \pm 0.10 ^b	7.54 \pm 0.23 ^a	7.70 \pm 0.26 ^a	3.85 \pm 0.08 ^c	3.92 \pm 0.11 ^c
Ash	1.34 \pm 0.11 ^a	1.37 \pm 0.10 ^a	1.36 \pm 0.08 ^a	1.40 \pm 0.17 ^a	1.38 \pm 0.13 ^a

Values expressed as mean value \pm standard error. Different superscripts in a line represent statistical differences (p < 0.05)

Table 2 demonstrates the amino acid compositions of golden grey mullet (*Chelon auratus*) and flathead grey mullet (*Mugil cephalus*). Total amino acid contents were found between 13.39 \pm 0.24-14.20 \pm 0.15 g/100g in the muscle tissue of *C. auratus* and 13.45 \pm 0.20-14.08 \pm 0.24 g/100g in *M. cephalus*. The highest total amino acid content in both species was found in the winter and spring (p < 0.05). The amino acid compositions of the culture forms of

both species were statistically similar to the summer and autumn samples (p > 0.05). All essential amino acids were detected in all samples. Glutamine, asparagine, alanine, leucine and isoleucine were the highest-detected amino acids in all groups. Tryptophan was the lowest (p < 0.05). In muscle tissue of *C. auratus*, asparagine, glutamine, aspartate, serin, glutamate, histidine, alanine, tyrosine, cysteine, tryptophan and phenylalanine were found

statistically the same in all seasons and culture form ($p > 0.05$). Glycine, threonine and isoleucine were highest in the winter, while lysine was highest in the spring ($p < 0.05$). Valine, methionine and leucine were found to be the most common in both the winter and spring seasons. In *M. cephalus*, asparagine, glutamine, aspartate, serin, glycine, valine, methionine, leucine and lysine were statistically the same in all groups ($p > 0.05$). Glutamine was highest in the spring, while histidine was highest in the summer. Threonine and isoleucine were higher in the winter and spring seasons ($p < 0.05$). In culture forms of *C. auratus*, amino acids were statistically similar compared to the autumn and summer seasons ($p > 0.05$). No statistical differences were found in the total amino acid compositions of the culture form of *M. cephalus* ($p > 0.05$).

The digestible amino acid composition and DIAAS score of the grey mullet species are shown in Table 3. According to the results, only two ileal amino acids, valine and histidine, were found below the recommended daily intake ratios in some groups, which defines limiting amino acids for DIAAS calculation. In *C. auratus*, valine was found below the daily recommendations in all groups for young children. At the same time, it was also below the recommended levels in the autumn, summer and culture groups for adult reference patterns. Histidine was found below the recommended daily intake for young children in the winter and spring. In *M. cephalus*, histidine was found below the recommendations for young children only in winter and spring groups, similar to *C. auratus*. In contrast, valine was found in autumn and culture form. Notably, the amino acid compositions of *M. cephalus* were higher than the daily recommendations for adults in terms of all digestible ileal amino acids. DIAAS score of *C. auratus* ranged between 89-92 for young children and 98-103 for adults. In *M. cephalus*, the DIAAS score ranged between 93 and 102 for young children, whereas it was calculated between 100-112 for adults. It is clear that *M. cephalus* is richer in terms of amino acid quality than *C. auratus*, with the highest DIAAS scores.

The fatty acid compositions of grey mullets are presented in Table 4. The total saturated fatty acid value was found to be the lowest in the winter, with $39.67 \pm 0.37\%$ in total, while it was the highest in the summer season, with $43.53 \pm 0.41\%$ in *C. auratus*. Palmitic acid (C16:0) was the highest saturated fatty acid in all samples, followed by pentadecanoic (C15:0) and stearic acids (C18:0). In contrast, the lowest total saturated fatty acids content was detected in the spring, while the highest values were found in both the summer and autumn seasons in *M. cephalus*. The highest amount of saturated fatty acid found in the muscle tissues of both mullet species was palmitic acid ($p < 0.05$). It was followed by pentadecanoic acid (C15:0), myristic acid (C14:0) and stearic acids (C18:0). The total monounsaturated fatty acids were statistically similar within each group of both fish species ($p > 0.05$). The highest monounsaturated fatty acid was oleic acid (C18:1), followed by palmitoleic acid (C16:1) in all groups ($p < 0.05$). The amount of oleic acid did not change throughout the year or in the culture form of *C. auratus* ($p > 0.05$).

However, the oleic acid content was increased in culture-adapted *M. cephalus* ($p < 0.05$). The highest total polyunsaturated fatty acid values were found in the winter and spring seasons in both species. The highest polyunsaturated fatty acids were docosahexaenoic acid (DHA) (C22:6) and eicosapentaenoic acid (EPA) (C20:5) in *C. auratus*. In *M. cephalus*, DHA was the highest PUFA, followed by linoleic acid (C18:2) ($p < 0.05$). In *C. auratus*, DHA was detected at the highest in the winter ($p < 0.05$), while it was found statistically the same in other groups ($p > 0.05$). However, in *M. cephalus*, DHA was detected as the lowest in the cultured form ($p < 0.05$), even though no statistical differences were detected between seasons ($p > 0.05$). EPA was found to be highest in the winter and spring groups of *C. auratus* ($p < 0.05$); there were no statistical differences ($p > 0.05$) between groups in *M. cephalus*. Essential fatty acids, linoleic acid (C18:2) and α -linolenic acid (C18:3) were detected in all groups. Linoleic acid was found statistically the same in both species' cultured forms and autumn, winter and spring seasons ($p > 0.05$). Only the summer values were found to be statistically different ($p < 0.05$) from the lowest. Similarly, α -linolenic acid was found to be the lowest in the summer and autumn samples ($p < 0.05$). *M. cephalus* had more total polyunsaturated fatty acids, DHA, linoleic acid and α -linolenic acid in all seasons and cultured forms than wild and cultured *C. auratus* (Table 4).

Lipid quality changes of grey mullet species are shown in Figure 3 and Table 5. According to the results, the highest PUFA/SFA ratios were detected in the winter and spring seasons in each species, but *M. cephalus* has higher values amongst species with 0.93 ± 0.04 and 0.94 ± 0.05 ($p < 0.05$). The highest FLQ of *C. auratus* groups was calculated in the winter season at 20.05 ± 0.35 , while the highest FLQ was found in the spring season of wild *M. cephalus* ($p < 0.05$). HPI and UI reached the highest proportions in the winter in *C. auratus* ($p < 0.05$). Similarly, in *M. cephalus*, UI was found to be highest in the winter along with the spring season ($p < 0.05$). However, HPI and HH were found to be statistically the same ($p > 0.05$). AI and TI were found to have the highest values in the summer in *C. auratus* ($p < 0.05$). Moreover, no statistical differences were detected in the LA/ALA, n3/n6, and NVI indices of *C. auratus* meat ($p > 0.05$). Similarly, in *M. cephalus*, seasonal values of LA/ALA, n3/n6, NVI, AI and TI indices were not statistically different ($p > 0.05$). In *C. auratus*, AI and TI were detected as the highest in the summer season ($p < 0.05$).

Table 2. Amino acid composition of wild and cultured forms of grey mullet species (g/100g)

	Golden grey mullet (<i>Chelon auratus</i>)					Flathead grey mullet (<i>Mugil cephalus</i>)				
	Autumn	Winter	Spring	Summer	Aquaculture	Autumn	Winter	Spring	Summer	Aquaculture
Asp	1.45±0.05 ^a	1.50±0.03 ^a	1.48±0.03 ^a	1.42±0.05 ^a	1.44±0.06 ^a	1.45±0.06 ^A	1.53±0.05 ^A	1.54±0.03 ^A	1.48±0.04 ^A	1.49±0.05 ^A
Glu	1.96±0.06 ^a	2.05±0.06 ^a	1.98±0.06 ^a	1.94±0.06 ^a	1.95±0.06 ^a	1.97±0.20 ^A	2.10±0.13 ^A	2.07±0.18 ^A	1.95±0.10 ^A	2.02±0.09 ^A
Asn	0.43±0.04 ^a	0.42±0.05 ^a	0.45±0.02 ^a	0.44±0.03 ^a	0.44±0.02 ^a	0.44±0.05 ^A	0.41±0.04 ^A	0.47±0.05 ^A	0.46±0.04 ^A	0.44±0.04 ^A
Ser	0.40±0.03 ^a	0.42±0.01 ^a	0.44±0.03 ^a	0.40±0.03 ^a	0.41±0.01 ^a	0.51±0.02 ^A	0.48±0.05 ^A	0.47±0.05 ^A	0.52±0.03 ^A	0.53±0.03 ^A
Gln	0.31±0.04 ^a	0.35±0.03 ^a	0.33±0.02 ^a	0.32±0.03 ^a	0.32±0.02 ^a	0.29±0.02 ^B	0.31±0.02 ^{AB}	0.34±0.02 ^A	0.28±0.01 ^B	0.31±0.02 ^{AB}
His	0.40±0.04 ^a	0.38±0.04 ^a	0.39±0.03 ^a	0.42±0.04 ^a	0.43±0.02 ^a	0.42±0.02 ^{AB}	0.35±0.03 ^B	0.38±0.03 ^{AB}	0.46±0.04 ^A	0.40±0.02 ^{AB}
Gly	0.53±0.02 ^b	0.58±0.02 ^a	0.53±0.03 ^b	0.51±0.03 ^b	0.52±0.02 ^b	0.55±0.04 ^A	0.51±0.05 ^A	0.50±0.04 ^A	0.54±0.02 ^A	0.58±0.03 ^A
Thr	0.69±0.03 ^{ab}	0.75±0.04 ^a	0.71±0.03 ^{ab}	0.64±0.02 ^b	0.65±0.04 ^b	0.64±0.03 ^B	0.72±0.04 ^A	0.74±0.02 ^A	0.63±0.03 ^B	0.66±0.03 ^B
Ala	1.39±0.06 ^a	1.48±0.04 ^a	1.42±0.06 ^a	1.36±0.05 ^a	1.35±0.07 ^a	1.38±0.06 ^A	1.43±0.05 ^A	1.40±0.04 ^A	1.35±0.05 ^A	1.41±0.06 ^A
Try	0.49±0.05 ^a	0.46±0.05 ^a	0.49±0.04 ^a	0.50±0.04 ^a	0.51±0.04 ^a	0.50±0.02 ^A	0.53±0.03 ^A	0.51±0.02 ^A	0.49±0.03 ^A	0.51±0.03 ^A
Cys	0.17±0.03 ^a	0.19±0.03 ^a	0.15±0.02 ^a	0.16±0.02 ^a	0.20±0.03 ^a	0.16±0.02 ^A	0.16±0.02 ^A	0.18±0.02 ^A	0.13±0.04 ^A	0.18±0.03 ^A
Val	0.72±0.03 ^b	0.81±0.04 ^a	0.83±0.04 ^a	0.75±0.04 ^b	0.74±0.02 ^b	0.75±0.05 ^A	0.79±0.03 ^A	0.82±0.04 ^A	0.79±0.03 ^A	0.77±0.04 ^A
Met	0.47±0.02 ^b	0.54±0.03 ^a	0.55±0.03 ^a	0.48±0.02 ^b	0.51±0.02 ^{ab}	0.48±0.03 ^A	0.54±0.05 ^A	0.50±0.03 ^A	0.49±0.04 ^A	0.51±0.02 ^A
Trp	0.15±0.01 ^a	0.17±0.01 ^a	0.16±0.01 ^a	0.16±0.01 ^a	0.18±0.01 ^a	0.12±0.01 ^A	0.13±0.01 ^A	0.14±0.01 ^A	0.12±0.01 ^A	0.13±0.01 ^A
Phe	0.63±0.07 ^a	0.65±0.06 ^a	0.66±0.04 ^a	0.60±0.06 ^a	0.61±0.02 ^a	0.58±0.04 ^A	0.60±0.04 ^A	0.61±0.03 ^A	0.59±0.04 ^A	0.62±0.03 ^A
Ile	0.75±0.03 ^{ab}	0.80±0.03 ^a	0.77±0.03 ^{ab}	0.72±0.02 ^b	0.73±0.04 ^b	0.72±0.02 ^B	0.75±0.03 ^A	0.76±0.02 ^A	0.70±0.03 ^B	0.71±0.01 ^B
Leu	1.32±0.03 ^b	1.40±0.04 ^a	1.42±0.04 ^a	1.31±0.03 ^b	1.35±0.02 ^{ab}	1.29±0.08 ^A	1.36±0.06 ^A	1.37±0.06 ^A	1.30±0.07 ^A	1.32±0.10 ^A
Lys	1.19±0.04 ^c	1.25±0.03 ^b	1.33±0.04 ^a	1.26±0.02 ^b	1.22±0.03 ^{bc}	1.20±0.07 ^A	1.27±0.11 ^A	1.28±0.08 ^A	1.24±0.06 ^A	1.22±0.07 ^A
∑AA	13.45±0.23 ^b	14.20±0.15 ^a	14.09±0.21 ^a	13.39±0.24 ^b	13.56±0.23 ^b	13.45±0.20 ^B	13.97±0.18 ^A	14.08±0.24 ^A	13.52±0.18 ^B	13.81±0.21 ^{AB}

Asp: aspartic acid, Glu: glutamic acid, Asn: asparagine, Ser: serine, Gln: glutamine, His: histidine, Gly: glycine, Thr: threonine, Ala: alanine, Tyr: tyrosine, Cys: cysteine, Val: valine, Met: methionine, Trp: tryptophan, Phe: phenylalanine, Ile: isoleucine, Leu: leucine, Lys: lysine, ∑AA: total amino acids. Values are expressed as mean value±standard error. Different superscripts in a line represent statistical differences. Sentenced case letters defined statistical differences between golden grey mullets, while capital letters were used for flathead grey mullets. The p-value is accepted as 0.05.

Table 3. Digestible indispensable amino acid ratio (DIAA) and scores (DIAAS) for wild and culture forms of grey mullet species

DIAA calculations			Golden grey mullet (<i>Chelon auratus</i>)					Flathead grey mullet (<i>Mugil cephalus</i>)				
Reference pattern	Amino acids	Daily intake	Autumn	Winter	Spring	Summer	Aquaculture	Autumn	Winter	Spring	Summer	Aquaculture
For young children (6 months to 3 years)	AAA	52	1.11	1.04	1.02	1.06	1.09	1.07	1.14	1.13	1.10	1.15
	His	20	1.02	0.92	0.89	1.04	1.07	1.06	0.90	0.98	1.20	1.04
	Ile	32	1.30	1.32	1.20	1.22	1.24	1.24	1.32	1.34	1.25	1.27
	Leu	66	1.09	1.10	1.05	1.05	1.09	1.06	1.14	1.14	1.10	1.12
	Lys	57	1.16	1.16	1.17	1.20	1.17	1.16	1.26	1.27	1.25	1.22
	SAA	27	1.28	1.38	1.25	1.25	1.39	1.27	1.41	1.37	1.27	1.41
	Thr	31	1.27	1.31	1.17	1.14	1.17	1.17	1.34	1.37	1.19	1.24
	Trp	8.5	1.01	1.08	0.97	1.05	1.18	1.01	1.09	1.16	1.04	1.10
	Val	43	0.90	0.96	0.93	0.92	0.91	0.93	1.00	1.04	1.02	0.99
	DIAAS %			90	92	89	92	91	93	90	98	102
Limiting AA			Val	His	His	Val	Val	Val	His	His	Val	Val
For older children, adolescents and adults	AAA	41	1.41	1.32	1.30	1.35	1.38	1.35	1.44	1.43	1.40	1.46
	His	16	1.27	1.15	1.11	1.30	1.34	1.33	1.13	1.22	1.50	1.30
	Ile	32	1.39	1.41	1.28	1.30	1.33	1.33	1.41	1.43	1.34	1.35
	Leu	61	1.18	1.19	1.14	1.14	1.18	1.14	1.23	1.24	1.19	1.21
	Lys	48	1.38	1.38	1.38	1.43	1.39	1.38	1.49	1.50	1.48	1.45
	SAA	23	1.50	1.62	1.47	1.46	1.63	1.49	1.66	1.61	1.49	1.66
	Thr	25	1.57	1.62	1.45	1.42	1.45	1.45	1.66	1.70	1.47	1.54
	Trp	6.6	1.31	1.39	1.24	1.35	1.52	1.30	1.40	1.49	1.34	1.42
	Val	40	0.97	1.03	1.00	0.99	0.98	1.00	1.08	1.12	1.09	1.06
	DIAAS %			97	103	100	99	98	100	108	112	109
Limiting AA			Val	Val	Val	Val	Val	Val	Val	Val	Val	Val

AAA: aromatic amino acids (phenylalanine + tyrosine), His: histidine, Ile: isoleucine, Leu: leucine, Lys: lysine, SAA: sulfur amino acids (methionine + cystine), Thr: threonine, Trp: tryptophan, Val: valine. IAA references for young children and adults were obtained from FAO (2013) and daily intakes were expressed as mg amino acid/g protein.

Table 4. Fatty acid composition of wild and cultured forms of grey mullet species (%)

Fatty Acids	Golden grey mullet (<i>Chelon auratus</i>)					Flathead grey mullet (<i>Mugil cephalus</i>)				
	Autumn	Winter	Spring	Summer	Aquaculture	Autumn	Winter	Spring	Summer	Aquaculture
C _{10:0}	0.11±0.02 ^a	0.02±0.01 ^b	0.10±0.02 ^a	0.08±0.01 ^a	0.07±0.02 ^a	0.08±0.02 ^B	0.13±0.02 ^A	0.07±0.01 ^B	0.13±0.02 ^A	0.14±0.02 ^A
C _{12:0}	0.22±0.02 ^a	0.18±0.03 ^{ab}	0.19±0.02 ^{ab}	0.18±0.03 ^{ab}	0.15±0.02 ^b	0.32±0.03 ^A	0.29±0.03 ^A	0.26±0.04 ^A	0.34±0.04 ^A	0.28±0.04 ^A
C _{14:0}	3.01±0.13 ^{bc}	2.35±0.12 ^d	3.22±0.07 ^b	4.25±0.08 ^a	2.87±0.11 ^c	2.91±0.14 ^A	2.82±0.10 ^A	2.70±0.18 ^{AB}	2.65±0.14 ^{AB}	2.60±0.10 ^B
C _{15:0}	8.31±0.12 ^b	7.54±0.13 ^d	7.97±0.09 ^c	8.63±0.18 ^a	8.34±0.13 ^b	8.31±0.25 ^A	7.65±0.26 ^B	7.75±0.19 ^B	8.45±0.31 ^A	8.01±0.14 ^{AB}
C _{16:0}	24.40±0.31 ^a	23.65±0.30 ^c	23.30±0.24 ^b	24.61±0.38 ^a	24.42±0.23 ^a	22.40±0.17 ^A	22.05±0.32 ^{AB}	21.86±0.30 ^B	22.65±0.37 ^A	22.34±0.15 ^A
C _{17:0}	1.56±0.09 ^b	1.59±0.08 ^b	1.61±0.11 ^b	1.88±0.12 ^a	1.86±0.15 ^a	1.92±0.07 ^A	1.66±0.10 ^B	1.48±0.07 ^C	1.90±0.15 ^A	1.91±0.11 ^A
C _{18:0}	3.24±0.10 ^{ab}	3.66±0.08 ^a	3.20±0.15 ^{ab}	2.88±0.22 ^b	3.31±0.23 ^{ab}	3.30±0.11 ^A	3.28±0.12 ^A	3.20±0.08 ^{AB}	3.21±0.10 ^{AB}	3.06±0.08 ^B
C _{20:0}	0.25±0.03 ^b	0.18±0.02 ^c	0.22±0.02 ^{bc}	0.29±0.02 ^a	0.23±0.03 ^{bc}	0.52±0.05 ^B	0.53±0.03 ^B	0.54±0.02 ^B	0.66±0.04 ^A	0.64±0.04 ^A
C _{21:0}	0.26±0.03 ^c	0.20±0.02 ^d	0.35±0.04 ^b	0.44±0.04 ^a	0.35±0.04 ^b	0.66±0.03 ^{AB}	0.60±0.03 ^B	0.61±0.04 ^B	0.65±0.03 ^{AB}	0.71±0.03 ^A
C _{23:0}	0.33±0.03 ^a	0.30±0.02 ^a	0.32±0.03 ^a	0.29±0.03 ^a	0.20±0.03 ^b	0.52±0.04 ^A	0.51±0.03 ^A	0.56±0.04 ^A	0.52±0.05 ^A	0.56±0.03 ^A
Σ_{SFA}	41.69±0.34^b	39.67±0.37^d	40.48±0.43^c	43.53±0.41^a	41.80±0.36^b	40.94±0.44^A	39.52±0.43^C	39.03±0.47^D	41.16±0.34^A	40.25±0.24^B
C _{14:1}	0.19±0.02 ^{ab}	0.16±0.021 ^b	0.21±0.02 ^a	0.20±0.02 ^a	0.18±0.01 ^{ab}	0.11±0.02 ^A	0.13±0.02 ^A	0.14±0.02 ^A	0.12±0.02 ^A	0.10±0.02 ^A
C _{15:1}	0.24±0.02 ^a	0.26±0.03 ^a	0.24±0.01 ^a	0.24±0.02 ^a	0.19±0.03 ^a	0.32±0.03 ^{AB}	0.31±0.02 ^{AB}	0.33±0.02 ^{AB}	0.36±0.02 ^A	0.27±0.02 ^B
C _{16:1}	8.11±0.17 ^c	8.86±0.12 ^a	8.39±0.11 ^b	8.04±0.09 ^c	8.62±0.20 ^a	7.13±0.21 ^B	7.32±0.18 ^{AB}	7.56±0.17 ^A	7.06±0.16 ^B	7.58±0.08 ^A
C _{17:1}	2.98±0.08 ^a	2.91±0.14 ^a	2.88±0.13 ^a	2.11±0.09 ^a	2.06±0.12 ^b	2.75±0.12 ^{AB}	2.65±0.07 ^B	2.89±0.15 ^A	2.21±0.14 ^C	2.34±0.13 ^C
C _{18:1}	12.68±0.31 ^a	12.41±0.32 ^a	12.58±0.34 ^a	12.74±0.35 ^a	12.92±0.32 ^a	10.36±0.23 ^B	10.28±0.40 ^B	10.50±0.15 ^B	10.67±0.21 ^{AB}	10.99±0.24 ^A
C _{20:1}	1.75±0.08 ^a	1.71±0.06 ^a	1.74±0.05 ^a	1.80±0.10 ^a	1.76±0.09 ^a	1.75±0.14 ^A	1.70±0.12 ^A	1.63±0.15 ^A	1.73±0.13 ^A	1.80±0.10 ^A

Table 4 continued

C _{22:1}	1.44±0.11 ^a	1.41±0.16 ^a	1.43±0.07 ^a	1.43±0.12 ^a	1.40±0.13 ^a	1.50±0.12 ^A	1.58±0.16 ^A	1.46±0.11 ^A	1.50±0.13 ^A	1.53±0.14 ^A
∑MUFA	27.39±0.18^{ab}	27.72±0.25^a	27.47±0.17^{ab}	26.56±0.36^b	27.13±0.27^{ab}	23.92±0.31^{AB}	23.97±0.38^{AB}	24.51±0.27^A	23.65±0.28^B	24.61±0.32^A
C _{18:2}	5.61±0.12 ^a	5.84±0.13 ^a	5.59±0.12 ^a	5.11±0.15 ^b	5.46±0.15 ^a	6.15±0.20 ^{AB}	6.42±0.19 ^A	6.35±0.13 ^A	6.00±0.20 ^B	6.20±0.18 ^{AB}
C _{18:3}	2.21±0.08 ^b	2.31±0.07 ^{ab}	2.40±0.05 ^a	2.08±0.06 ^c	2.28±0.08 ^{ab}	3.39±0.12 ^C	3.89±0.08 ^A	3.67±0.11 ^B	3.34±0.11 ^C	3.64±0.09 ^B
C _{20:2}	0.94±0.05 ^a	0.80±0.07 ^b	0.91±0.06 ^a	0.85±0.05 ^{ab}	0.91±0.04 ^a	1.21±0.15 ^A	1.29±0.14 ^A	1.25±0.07 ^A	1.14±0.17 ^A	1.16±0.10 ^A
C _{20:3}	0.88±0.04 ^a	0.89±0.03 ^a	0.92±0.05 ^a	0.83±0.05 ^a	0.86±0.05 ^a	1.64±0.15 ^A	1.68±0.16 ^A	1.64±0.17 ^A	1.59±0.15 ^A	1.57±0.13 ^A
C _{20:4}	1.12±0.06 ^a	1.21±0.05 ^a	1.15±0.06 ^a	1.18±0.07 ^a	1.19±0.06 ^a	1.35±0.17 ^A	1.36±0.21 ^A	1.38±0.20 ^A	1.38±0.18 ^A	1.37±0.14 ^A
C _{20:5}	6.29±0.29 ^b	7.23±0.27 ^a	7.01±0.18 ^a	6.48±0.28 ^b	6.87±0.19 ^{ab}	5.89±0.24 ^A	6.01±0.26 ^A	6.28±0.27 ^A	6.12±0.21 ^A	6.20±0.26 ^A
C _{22:5}	1.52±0.13 ^a	1.51±0.11 ^a	1.56±0.15 ^a	1.30±0.09 ^b	1.40±0.15 ^b	1.36±0.16 ^A	1.46±0.19 ^A	1.55±0.18 ^A	1.43±0.16 ^A	1.40±0.15 ^A
C _{22:6}	12.23±0.23 ^b	12.82±0.27 ^a	12.48±0.28 ^b	12.06±0.31 ^b	12.06±0.27 ^b	14.12±0.25 ^A	14.36±0.27 ^A	14.35±0.26 ^A	14.09±0.24 ^A	13.56±0.18 ^B
∑PUFA	30.80±0.41^b	32.61±0.44^a	32.02±0.40^a	29.89±0.47^c	31.03±0.51^b	35.11±0.35^B	36.47±0.38^A	36.47±0.41^A	35.09±0.29^B	35.10±0.32^B

∑SFA total saturated fatty acids, ∑MUFA total monounsaturated fatty acids, ∑PUFA total polyunsaturated fatty acids, C10:0 capric acid, C12:0 lauric acid, C14:0 myristic acid, C15:0 pentadecanoic acid, C16:0 palmitic acid, C17:0 margaric acid, C18:0 stearic acid, C20:0 arachidic acid, C21:0 heneicosylic acid, C23:0 Tricosanoic acid, C14:1 myristoleic acid, C15:1 pentadecenoic acid, C16:1 palmitoleic acid, C17:1 heptadecenoic acid, C18:1 oleic acid, C20:1 eicosenoic acid, C22:1 erucic acid, C18:2 linoleic acid, C18:3 α -linolenic acid, C20:2 eicosadienoic acid, C20:3 dihomogamma linolenic acid, C20:4 arachidonic acid, C20:5 eicosapentaenoic acid (EPA), C22:5 docosapentaenoic acid, C22:6 docosahexaenoic acid (DHA). Values are expressed as mean value±standard error. Different superscripts in a line represent statistical differences. Sentenced case letters defined statistical differences between golden grey mullets, while capital letters were used for flathead grey mullets. The p-value is accepted as 0.05.

Table 5. Lipid quality indices of wild and cultured forms of grey mullet species

	Golden grey mullet (<i>Chelon auratus</i>)					Flathead grey mullet (<i>Mugil cephalus</i>)				
	Autumn	Winter	Spring	Summer	Aquaculture	Autumn	Winter	Spring	Summer	Aquaculture
EPA+DHA	511.1±20.88 ^c	1457.6±42.65 ^a	1239.6±37.25 ^b	391.2±19.73 ^d	422.1±15.24 ^d	1034.5±31.32 ^B	1535.9±40.55 ^A	1588.5±37.64 ^A	778.1±25.44 ^C	774.6±24.19 ^C
PUFA/SFA	0.74±0.03 ^{ab}	0.82±0.05 ^a	0.79±0.03 ^a	0.69±0.04 ^b	0.74±0.03 ^{ab}	0.86±0.05 ^{AB}	0.93±0.04 ^A	0.94±0.05 ^A	0.85±0.03 ^B	0.87±0.04 ^{AB}
LA/ALA	2.54±0.11 ^a	2.53±0.09 ^a	2.33±0.13 ^a	2.46±0.09 ^a	2.39±0.12 ^a	1.81±0.15 ^A	1.65±0.06 ^A	1.73±0.12 ^A	1.80±0.05 ^A	1.70±0.09 ^A
n-3/n-6	3.02±0.09 ^a	3.15±0.13 ^a	3.19±0.07 ^a	3.19±0.08 ^a	3.10±0.14 ^a	3.03±0.18 ^A	3.02±0.15 ^A	3.06±0.19 ^A	3.12±0.26 ^A	3.02±0.09 ^A
NVI	0.65±0.05 ^a	0.68±0.04 ^a	0.68±0.07 ^a	0.63±0.03 ^a	0.66±0.04 ^a	0.61±0.02 ^A	0.61±0.03 ^A	0.63±0.02 ^A	0.61±0.04 ^A	0.63±0.02 ^A
FLQ	18.54±0.32 ^b	20.05±0.35 ^a	19.50±0.36 ^{ab}	18.54±0.38 ^b	18.94±0.30 ^b	20.02±0.26 ^B	20.38±0.28 ^{AB}	20.63±0.30 ^A	20.23±0.22 ^{AB}	19.77±0.31 ^B
HPI	1.59±0.08 ^b	1.82±0.07 ^a	1.64±0.05 ^b	1.35±0.07 ^c	1.61±0.09 ^b	1.72±0.09 ^A	1.80±0.13 ^A	1.85±0.08 ^A	1.75±0.10 ^A	1.81±0.07 ^A
UI	166.67±1.54 ^c	176.06±1.82 ^a	172.76±2.06 ^b	163.19±1.35 ^d	167.76±1.72 ^c	180.10±2.15 ^B	185.05±2.33 ^A	186.41±2.45 ^A	180.53±2.04 ^B	179.80±2.30 ^B
h/H	1.59±0.04 ^b	1.73±0.05 ^a	1.68±0.06 ^{ab}	1.48±0.06 ^c	1.61±0.04 ^b	1.80±0.13 ^A	1.88±0.08 ^A	1.91±0.09 ^A	1.81±0.11 ^A	1.85±0.11 ^A
AI	0.63±0.04 ^b	0.55±0.03 ^c	0.61±0.03 ^{bc}	0.74±0.05 ^a	0.62±0.02 ^b	0.58±0.03 ^A	0.56±0.04 ^A	0.54±0.04 ^A	0.57±0.02 ^A	0.55±0.03 ^A
TI	1.12±0.05 ^{ab}	1.05±0.05 ^b	1.06±0.04 ^b	1.18±0.05 ^a	1.11±0.06 ^{ab}	1.05±0.05 ^A	1.01±0.03 ^A	0.99±0.03 ^A	1.05±0.04 ^A	1.01±0.04 ^A

EPA: eicosapentaenoic acid, DHA: docosahexaenoic acid, PUFA: polyunsaturated fatty acids, SFA: saturated fatty acids, LA: linoleic acid, ALA: alpha-linolenic acid, NVI: nutritive value index, FLQ: fish lipid quality, HPI: health promoting index, UI: unsaturation index, h/H: hypo/hypercholesterolemic ratio, AI: atherogenicity index, TI: thrombogenicity index. Values are expressed as mean value±standard error. Different superscripts in a line represent statistical differences. Sentenced case letters defined statistical differences between golden grey mullets, while capital letters were used for flathead grey mullets. The p-value is accepted as 0.05.

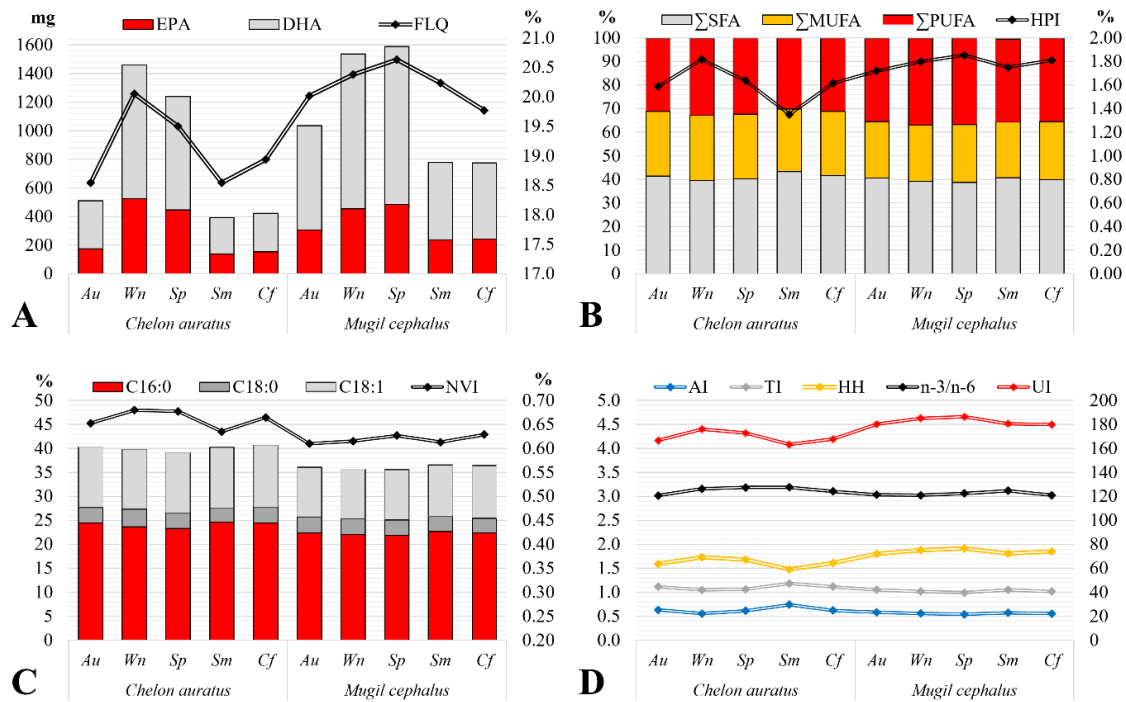


Figure 3. Changes in lipid quality. A: Food lipid quality (FLQ) with EPA and DHA, B: Health-promoting index (HPI), C: Nutritive value index (NVI), D: Indices of atherogenicity (AI), thrombogenicity (TI), unsaturation (UI), hypo/hypercholesterolemia (HH) and n-3/n-6 ratio

Discussion

Seafood contains high levels of essential amino acids and polyunsaturated fatty acids necessary for human nutrition (Özden and Erkan, 2008; Sahena et al., 2009). These nutrients are used for many essential tasks, such as maintaining metabolic activities, repairing organs and tissues, and regulating hormones and secretions (Ballantyne, 2011). Aquaculture is one of the main ways to meet the increasing global food demand with nutritious products like seafood (FAO, 2020). The food quality of aquaculture-derived seafood varies depending on many factors, such as nutrition, season, size, age, and water temperature (Nurnadia et al., 2011; Bjorndal and Guillen, 2016). In this research, both species show similar changes in proximate composition by the seasons. Crude fat content, in particular, increases in the winter season when water temperatures drop and decreases in the summer and autumn seasons in parallel with the reproduction period of the fish. The differences in meat quality are believed to be mainly a factor of seasonal and reproductive patterns of the fish (Di Lena et al., 2016). Besides, fish adapted to culture conditions were sampled at the end of one year. Environmental conditions were constant during sampling. Sampling was carried out in May 2018, and the salinity of the water used during the sampling was 0.18‰, the temperature was 15 °C, the nitrite nitrogen value was 0.046 MM, the nitrate nitrogen value was 1.312, the ammonia value was 0.001 MM, the phosphate value was 0.108 MM and silicon value 4.637 MM. Since the environmental conditions and nutritional characteristics were similar in the

study, the nutrient content of the fish in culture conditions was similar.

When the amino acid contents of two mullet species were compared with the individuals caught in nature, it was observed that the results obtained were statistically similar to the summer and autumn samples ($p > 0.05$). Although no statistical difference was detected between seasons for most amino acids ($p > 0.05$), statistical differences were found between total amino acid contents ($p < 0.05$). Statistically, the lowest total amino acid amounts were obtained in these periods. Aspartate and glutamate are the most abundant amino acids in tissues. In the pretreatment of amino acid analysis, fish meat is hydrolyzed with high temperature and low pH (Çankırılıgil et al., 2020). Due to this hydrolyzation process, asparagine and glutamine, which are intolerant to heat and low pH, are broken down into aspartate and glutamate (Varlık et al., 2004; Moreno et al., 2012). Similarly, tryptophan, which is highly sensitive to environmental conditions, is reduced during this acidic digestion process (Varlık et al., 2004). For this reason, tryptophan, asparagine and glutamine amounts were found to be lower, while aspartate and glutamate were found to be higher.

The DIAAS score defines the food's protein quality, and the highest values were calculated for *M. cephalus*. FAO (2013) states that a DIAAS score ranges between 75 - 99, which means the sample food can be accepted as a good protein source. If the DIAAS score is 100 or higher, the sample food is an excellent food source in terms of protein quality. According to the results, *M. cephalus* is an excellent protein source for older children, adolescents and adults in

all seasons. At the same time, it was an excellent protein source for young children (6 months to 3 years) in winter, spring and summer seasons, exceeding 100 points. *C. cephalus* was detected as an excellent protein source only for older children, adolescents and adults in the winter and spring.

Palmitic acid (C16:0) is often fish's most abundant saturated fatty acid (Yu et al., 2020). Comparing the fatty acid differences between the groups, it can be seen that the results are similar with crude nutrient composition and amino acid composition. Unsaturated fatty acids tend to increase in the winter, while saturated fatty acids tend to decrease. Similarly, the lowest amounts of polyunsaturated fatty acids were found in the summer. According to the literature, EPA (C20:5) and DHA (C22:6) are higher in pelagic fish in colder waters than in other fish (Hossain, 2011). As a result, the meat quality of both species varies mainly due to environmental conditions resulting from seasonal differences and reproductive periods. When we compared it to aquaculture-adapted individuals, it was shown that the cultured form of *M. cephalus* had more total polyunsaturated fatty acids, DHA (mg), than that in *C. auratus* (Table 4). However, the cultured form of *M. cephalus* had more lipid content than *C. auratus*, which was approximately 1.75 times higher. For this reason, although the proportional amount of EPA in the cultured form of *M. cephalus* was found to be less than that of *C. auratus* when proportioned with the crude fat amounts of the species, it is seen that *M. cephalus* contains more EPA with 243.04 mg than *C. auratus* as 153.20 mg. Similarly, when expressed in milligrams, the total amount of DHA in the cultured form of *M. cephalus* is approximately 1.97 times higher (531.55 mg) than *C. auratus* (268.94 mg). In similar studies on grey mullets, palmitic acid, oleic acid, EPA and DHA were found at similar ratios (Alpaslan et al., 2019; Ramoz-Judez et al., 2023). Our results are similar to those reported in the literature.

In the Mediterranean diet, food sources having high PUFA/SFA, n3/n6, HPI, FLQ and NVI were linked to positive health effects such as reducing cholesterol and preventing cardiovascular diseases (Hossain, 2011; Chen and Liu, 2020). PUFA/SFA ratios were highest in winter and spring seasons in both species due to those seasons' rich polyunsaturated fatty acid contents (Table 4). In addition, FLQ was higher in these seasons due to low SFA and high PUFA proportions (Figure 3A). Similarly, HPI was also found to be highest in the groups with more polyunsaturated fatty acids than saturated ones (Figure 3B). NVI was associated with palmitic acid (C16:0), stearic acid (C18:0) and oleic acid (C18:1) of the food source, and it was found higher in groups having relatively low palmitic acid and high oleic acid, such as winter and spring seasons of *C. auratus* (Figure 3C). UI is characterized by the amount of unsaturated fatty acids and double bond counts in each fatty acid (Chen and Liu, 2020). Thus, UI increased parallel with DHA and EPA contents, which have 6 and 5 double bonds, respectively. AI and TI indices were found to be inversely proportional to the h/H ratio (Figure 3D). Due to increasing some saturated fatty acids, such as lauric acid (C12:0),

myristic acid (C14:0), palmitic acid (C16:0) and stearic acid (C18:0), AI and TI can be high, but the h/H value is low. Similarly, as the amounts of unsaturated fatty acids increase, the h/H ratio decreases, while AI and TI values increase (Ulbricht and Southgate, 1991; Santos-Silva, 2002). All three indices are directly related to cardiovascular health and must be within specific ranges for an ideal diet (Hossain, 2011). For fish, these values are between 0.21-1.07 for AI, 0.14-0.87 for TI, and 0.87-4.83 for h/H (Chen and Liu, 2020). The values of AI and h/H determined in our study are consistent with the literature. AI was found to be slightly higher than the literature in the range of 1.05 ± 0.05 - 1.18 ± 0.05 for *C. auratus* and 0.99 ± 0.03 - 1.05 ± 0.05 for *M. cephalus*.

In conclusion, aquaculture products, such as those from golden grey mullet (*Chelon auratus*) and flathead grey mullet (*Mugil cephalus*), offer biologically rich sources of essential amino acids and polyunsaturated fatty acids vital for human nutrition. This study demonstrates that the meat quality of these species, including their crude nutrient composition and amino acid content, varies seasonally and is influenced by their reproductive cycles. Notably, crude fat content increases during colder months and decreases during the reproductive period in the summer and autumn. Environmental conditions and feeding regimes in aquaculture settings can mirror these natural variations, resulting in similar nutrient profiles between cultured and wild-caught specimens. The high levels of EPA and DHA, as well as other fatty acids found in these fish, underscore their nutritional value. Ultimately, the findings highlight the significant impact of seasonal and reproductive factors on the meat quality of cultured fish, affirming their importance as a nutritious and sustainable food source.

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Conflict of Interest

The author declares no conflict of interest

Author Contributions

Ekrem Cem Çankırılıgil and Ayça Altuntaş planned and designed the research and performed sampling studies, data collection and analysis. Ayça Altuntaş carried out culture adaptation studies and provided fish material from aquaculture. Ekrem Cem Çankırılıgil performed chemical analysis and manuscript writing. All authors discussed the results and contributed to the final manuscript.

Ethics Approval

All animal experiments were performed according to the ARRIVE guidelines and EU directive 2010/63/EU. The study was ethically approved with the ETİK-2017/2 code by the Ethical Committee of Animal Experiments of Central Fisheries Research Institute, Ministry of Agriculture and Forestry, Republic of Türkiye.

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