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A Preliminary Study on the Meat Yield, Nutritional Composition, Lipid Quality Indices, and Mineral and Heavy Metal Contents of Annular Seabream (*Diplodus annularis* Linnaeus, 1758) Caught in the Black Sea

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Research Article

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Abstract

In this study, the meat yield, nutritional composition (protein, fat, moisture, ash, carbohydrate), amino acid content, fatty acid composition, and mineral and heavy metal contents of annular seabream (*Diplodus annularis)* were investigated. Additionally, the lipid quality indices (LQI) of annular sea bream were examined. The meat yield of annular seabream was found to be 29.69%. Protein, fat, moisture, ash and carbohydrate values were 17.78 g/100g, 2.28 g/100g, 76.96 g/100g, 1.80 g/100g and 1.19 g/100g, respectively. The total amount of essential amino acids and non-essential amino acids were determined as 147.45 mg/g and 462.74 mg/g, respectively. Polyunsaturated fatty acids (PUFAs) were the predominant fatty acids (37.74%), followed by saturated fatty acids (SFAs) (34.85%) and monounsaturated fatty acids (MUFAs) (27.36%). Atherogenicity Index (AI), Thrombogenicity Index (TI) and Polyene Index (PI) from LQIs were calculated as 0.47, 0.35 and 0.91, respectively. The H/H (Hypocholesterolemic/hypercholesterolemic) ratio of annular seabream was 2.83 and FLQ (Fish Lipid Quality) was 14.48. The first three of the most abundant minerals in annular sea bream were potassium (K), phosphorus (P) and sodium (Na). The heavy metals Hg, Pb and Cd were found within the allowed limit values. As a result, the annular seabream meat was found to have low fat values and high protein content, and to contain essential amino acids, polyunsaturated fatty acids and important minerals. The current study is the first research conducted on the nutritional composition of annular sea bream in the Black Sea.

Keywords: Annular seabream, meat yield, nutritional composition, amino acid, fatty acids, lipid quality indices, mineral composition, heavy metals

Karadeniz'de Avlanan Isparoz Balığı (*Diplodus annularis* Linnaeus, 1758)'nın Et Verimi, Besin Kompozisyonu, Lipid Kalite Indeksleri, Mineral ve Ağır Metal İçerikleri Üzerine Bir Ön Çalışma

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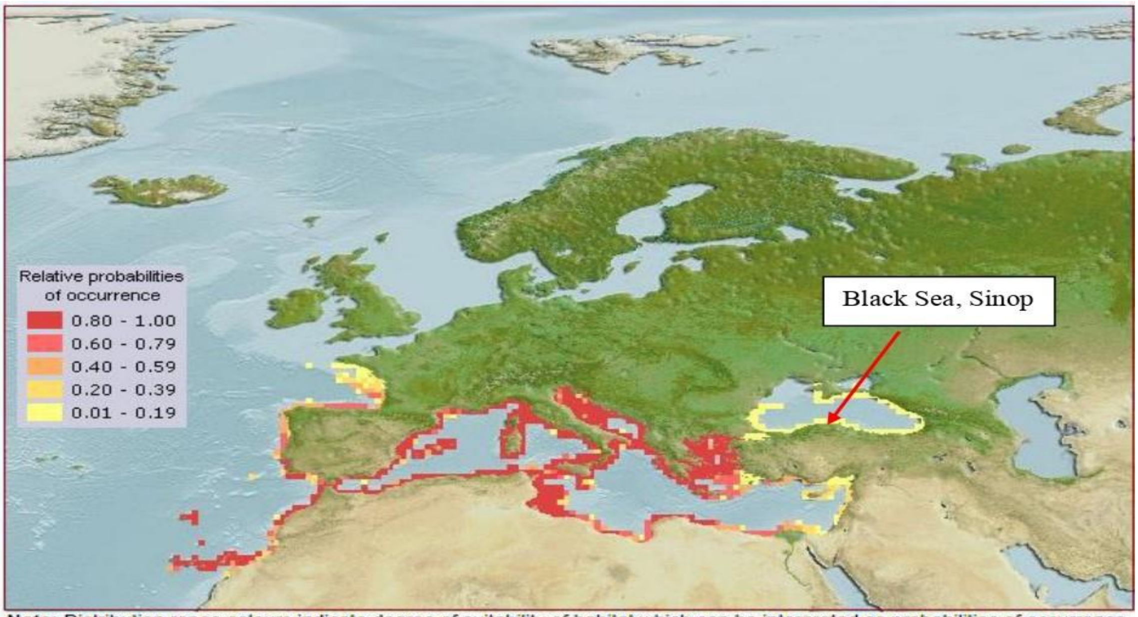
Öz

Bu çalışmada isparozun et verimi, besin kompozisyonu (protein, yağ, nem, kül, karbonhidrat), aminoasit içeriği, yağ asitleri kompozisyonu, mineral içeriği ve ağır metal içerikleri araştırılmıştır. Ayrıca isparoz balığının yağ kalite indeksleri incelenmiştir. Isparoz balığının et verimi %29.69 olarak bulunmuştur. Protein, yağ, nem, kül ve karbonhidrat değerleri sırasıyla, 17.78 g/100g, 2.28 g/100g, 76.96 g/100g, 1.80 g/100g ve 1.19 g/100g olarak tespit edilmiştir. Toplam esansiyel aminoasit miktarı 147.45 mg/g, toplam esansiyel olmayan aminoasit miktarı ise 462.74 mg/g olarak belirlenmiştir. Çoklu doymamış yağ asitleri (PUFAs) (37.74%) baskın yağ asitleri olup, onu doymuş yağ asitleri (SFAs) (34.85%) ve tekli doymamış yağ asitleri (MUFAs) (27.36%) takip etmiştir. Yağ kalite indekslerinden aterojenite indeksi (AI), trombojenite indeksi (TI) ve polien indeksi (PI) sırasıyla 0.47, 0.35 0.91 olarak tespit edilmiştir. İsparoz balığının H/H oranı 2.83 ve FLQ ise 14.48 olarak bulunmuştur. İsparoz balığında en fazla bulunan minerallerden ilk üçü potasyum (K), fosfor (P)ve sodyum (Na) olarak ölçülmüştür. Ağır metallerden Hg, Pb ve Cd izin verilen limit değerler içerisinde bulunmuştur. Sonuç olarak bu çalışmada isparoz balığı etinin, düşük yağ ve yüksek protein içeriğine sahip olduğu, esansiyel aminoasitleri, çoklu doymamış yağ asitlerini ve önemli mineralleri bünyesinde bulundurduğu tespit edilmiştir. Mevcut çalışma Karadeniz'de isparoz balığının besinsel kompozisyonuna yönelik olarak gerçekleştirilen ilk araştırma özelliğini taşımaktadır.

Anahtar Kelimeler: Isparoz, et verimi, besin kompozisyonu, aminoasit, yağ asitleri, yağ kalite indeksleri, mineral kompozisyonu, ağır metaller

Introduction

Fish is considered a nutritionally valuable part of the human diet and its consumption twice a week is recommended, generally due to the content of polyunsaturated fatty acids (PUFA) in fish meat [1]. Moreover, it is an important source of high-quality protein, valuable oils, vitamins and minerals [2]. The findings reveal that fish is a valuable source of essential amino acids and polyunsaturated fatty acids that play important physiological functions in the maintenance and development of fetuses, neonates, and infant brains [3]. For all these reasons, fish is a nutrient which human need beginning from the mother's womb till adulthood and throughout their lives. It is also an indispensable part of healthy and balanced nutrition. The annular seabream (*Diplodus annularis* Linnaeus, 1758) is a species of fish from the family Sparidae. It is locally known as 'ısparoz', 'ispari' or 'isparoz' in Türkiye. Its colours may be silver grey or yellow green depending on where it lives and what it eats. It is adapted to seagrasses in terms of appearance and colour. Its abdominal parts are generally white. The black band on the tail stalk is its most distinctive feature. Their bodies are covered with large scales. It is a demersal fish species. They survive in seagrass (*Posidonia*) beds on sandy bottoms and rarely on rocky bottoms. It is caught using trawl nets, bottom gillnets and fishing lines [4, 5]. In some regions, this species may prefer places where fish farms are located, to feed on fishmeal distributed from cages to the environment [6]. It is found in the Eastern Atlantic from southwestern France to southwestern Spain and Portugal, including the Canary Islands, throughout the Mediterranean, and in the Black Sea and the Sea of Azov [7]. The annular seabream can survive between depths of 0-90 meters. It feeds on worms, crustaceans, molluscs, echinoderms and hydrozoans. Although this is a hermaphrodite species, both sexes are seen in a population. Some individuals are protandric. Although the maturity length varies between 8-19.6 cm, the maximum growth length has been reported as 28 cm [7]. Despite the fact that the breeding time depends on water temperature, the periods were reported as June-August in the Black Sea, April-August in the Marmara Sea and the Mediterranean, and March-August in the Aegean Sea.



Note: Distribution range colours indicate degree of suitability of habitat which can be interpreted as probabilities of occurrence.

Figure 1. *Distribution of annular seabream (Diplodus annularis) in marine areas and the sampling area [7]*

Recommended consumption months of annular seabream are January, February, October, November and December for the Marmara Sea and the Mediterranean; January, February, March, April, October, November and December for the Black Sea; and January, October, November and December for the Aegean Sea [8]. The production quantity of annular sea bream over the past decade (2013-2022) has been reported as 106.6, 58.7, 75, 84.2, 86.6, 45.9, 54.1, 54, 55, 38.2 tonnes/per year, respectively [9]. When the available data is considered, it can be said that the production amount of annular seabream can be suggested to generally decrease over time. The population density of annular seabream in the Black Sea is lower than those at the Aegean and the Mediterranean coasts of Türkiye and in the European and African coasts of the Eastern Atlantic. The quite low production amount is probably due to the low population density on the Turkish coast, as well as the fact that hunting as a target species is not a much-preferred approach [6]. Many studies have been carried out on the age, growth, feeding habits and nets used in hunting the annular sea bream [10 – 14]. In addition, the species has been the subject of various studies in recent years to evaluate marine pollution and ecological risks [15, 16]. Studies on the chemical composition of annular sea bream are quite limited [17, 18]. Information on the fatty acid content of annular seabream can be obtained through literature [19 – 21]. Annular seabream is not an economically valuable fish. It is not caught as a target species. However, it is a by-catch, which can be sold by fishermen at certain periods in some provinces of the Black Sea, especially in Sinop. Research on the nutritional composition of this fish is quite limited. Moreover, no studies on the nutritional composition of annular sea bream caught in the Black Sea have been encountered in literature. The current study will serve as a pioneering work and is important in terms of its contribution to literature as well as its informative content to consumers. This study aims to examine the meat yield, nutritional composition, amino acids, fatty acids, lipid quality indices, and the mineral and heavy metal contents of annular seabream caught in the Black Sea. It is thought that the data obtained will provide significant contributions to future studies on the subject.

Materials and Methods

Material

In the present study, 2 kg (n=10) of annular seabream (*Diplodus annularis*) with average lengths of 19.04±0.24 cm and average weights of 136.30±6.02 g were used as raw material. To create homogeneous samples, larger fish were selected based on their sexual maturity. The fish were purchased from a local fisherman in Sinop, (Türkiye) in May 2023. The fish were covered with ice chips in styrofoam boxes and brought to the laboratory in 20 minutes.

Methods

Determination of Meat Yield (%)

The head, fins, internal organs, skin and bones of the fish were cut and weighed separately to calculate the meat yield of the fish. Following removal of the head, fins, skin, bones and all internal organs, the meat weight of the fish was assessed. The ratio of edible meat weight to total body weight was calculated as meat yield. The obtained values were used in the formula below and the results were evaluated.

Meat yield (%) = [edible meat weight (g)/whole weight(g)]x100 [22]

Nutritional Composition Analyses

Nutritional Composition

Crude protein and crude fat contents of the samples were determined according to the methods of AOAC [23] and Bligh and Dyer [24], respectively. Determination of % moisture content was carried out according to Ludorf and Meyer [25]. Crude ash contents were analyzed according to the methods used by AOAC [26]. Energy value was calculated following Falch et al. [27]. All measurements were carried out in triplicate.

Amino Acids

Homogenized fish meat (0.5 g) was weighed and burned with 20 ml HCl at 110°C for 18-24 hours [28]. 20 ml of pure water was added to the sample and it was dried in an evaporator at 70°C. The volume was completed to 50 ml in a volumetric flask with pure water. Amino acids were separated by Zorbax Eclipse AAA, UFLC using a 4.6 X 150 mm, 3.5 µm column. The flow rate was set to 1 mL/min and the column temperature was set to 40 °C. All samples were analyzed in duplicate and the results were given in mg/g.

Fatty Acids

Fatty acid compositions of samples were detected by Gas Chromatography/Mass Spectrometry (GC/MS, Thermo Scientific ISQ LT) equipped with an autosampler. The capillary column used was Trace Gold TG-WaxMS (60 m) with an inner diameter of 0.25 µm and a thickness of 0.25 µm. The temperature was held initially at 100°C for 3 min and then increased to 240°C at a rate of 4°C/min, with an initial hold of 6 min. The carrier gas used was helium (1 ml/min) and the split ratio was 1:20. Front inlet temperature was set to 240°C, MS transfer line temp and ion source temperatures were 250°C and 240°C, respectively. The mass spectrometer was operated in the electron impact ionization mode (70 eV). A FAME mix (Supelco, 37 comp., Bellefonte, PA, USA) was used to provide standards for comparison [29].

Lipid Quality Indices (LQI)

Atherogenic Index (AI), Thrombogenicity Index (TI), Polyene Index (PI), Hypocholesterolemic/hypercholesterolaemic Index (HH) and Fish Lipid Quality (FLQ) were calculated by using following equations described by Ulbricht and Southgate [30], Lubis and Buckle [31], Santos-Silva et al. [32] and Abrami et al. [33].

$$AI = \frac{\mathrm{C12+(4XC14)+C16}}{\mathrm{\Sigma n6+\Sigma n3+\Sigma MUFA}} \quad (1)$$

$$TI = \frac{\mathrm{C14+C16+C18}}{\mathrm{(0.5X\Sigma MUFA)+(0.5X\Sigma n{-}6)+(3x\Sigma n{-}3)+(n{-}3)/(n{-}6)}}. \quad (2)$$

$$PI = \frac{\mathrm{(C20{:}5+C22{:}6)}}{\mathrm{C16}}. \quad (3)$$

$$HH = \frac{(C18{:}1n{-}9 + C18{:}2n{-}6 + C18{:}3n{-}3 + C20{:}4n{-}6 + C20{:}5n{-}3 + C22{:}5n{-}3 + C22{:}6n{-}3)}{\mathrm{(C14{:}0 + C16{:}0)}}. \quad (4)$$

$$FLQ = \frac{\mathrm{(EPA\%+DHA\%)}}{\mathrm{Total\ fatty\ acids\%}} x100. \quad (5)$$

Determination of Minerals and Heavy Metals

According to the method described by Milestone [34] the acid (7 ml of HNO_3 65%, 1 ml H_2O_2 30%: Merck, Darmstadt, Germany) digestion of the sample in a closed vessel device is provided using a temperature control microwave (Ethos D, Milestone Inc. Sorisole, Italy) for the determination of metals by spectroscopic methods. Analyses of 28 elements (Macro, trace elements and heavy metals: Na, Mg, K, P, Ca, Si, Fe, Zn, Al, Rb, Sr, Cr, Ti, Be, Se, Cu, Mn, Li, Mo, Ni, Sb, Cs, Ba, Gd, As, Hg, Pb, Cd) were carried out using inductively coupled plasma mass spectrometry using dynamic reaction cell technology (Agilent Technologies / 7700X ICP-MS Systems). Results are expressed in milligrams of element per kilogram of fish meat. All samples were analyzed in triplicate.

Public Health Risk Analysis

Metal Pollution Index (MPI)

The following formula was used to find the MPI value;

$$MPI = (C1XC1X \dots \dots . XCn)\frac{1}{n} \quad (6)$$

Cn = metal concentration in the sample (mg·kg-1)
n = number of samples [35]

Estimated Daily Intake (EDI), Estimated Weekly Intake (EWI)

The weekly seafood consumption amount used in calculating the EDI and EWI values was calculated using the average per capita seafood consumption amount in Türkiye in 2022. This value is approximately 0.140 kg·week-1 (7.3 kg/52 weeks annually) [36].
The USEPA [37] equation (7) below was used to calculate the EDI value:

$$EDI = \frac{MxW}{BW} \quad (7)$$

where:
M = metal concentration in the sample (mg·kg-1)
W =daily amount of seafood consumption (kg)
BW = body weight (kg)
The relation between the EWI and EDI is presented in the equation (8).

$$EWI = EDIx7 \quad (8)$$

In the study, the average weight of an adult was taken as 70 kg (18 years old) and
32 kg for children (10 years old) [38].

Results and Discussion

Meat Yield (%)

The length, weight and meat yield of annular seabream are shown in Table 1. The average length and average weight of annular seabream were found as 19.04 cm and 136.30 g, respectively. Kasapoglu and Duzgunes [39] reported minimum and maximum length and weight values of annular sea bream in the Black Sea as 6.4-7.8 cm and 2.79-8.21 g. In another study conducted in the Black Sea, these values were given as 13.3-23 cm and 50.3-235.8 g [6]. In a study comparing the length and weight values of annular seabream for Turkish waters, the minimum/maximum length was reported as 12-23.2 cm and the weight as 28.69-180 g [13]. Samsun et al. [40] reported that the minimum and maximum length-weight values of annular seabream in the central Black Sea (Sinop) were 12.5-23.4 cm and 39.9-249.3 g, respectively.

Table 1. *The length, weight and meat yield of annular seabream (Diplodus annularis)*

	Min	Avg	Max
Fish Total Lenght (cm)	17.8	19.04±0.24	20
Fish Total Weight (g)	107.64	136.30±6.02	174.57
Fillet weight (g)	31.24	40.47±1.85	54.56
Meat yield (%)	27.33	29.69±0.58	33.38

Values are shown as mean ± standard error (n=10)

The average meat yield of annular seabream was found as 29.69%. To select homogeneous samples, larger fish were picked based on their sexual maturities. Lenght-weight ratios and meat yield % may vary depending on various factors. Age of the fish, catch region, maturity, nutritional status, water temperature, etc. are some of these factors.

Nutritional Composition

Protein, fat, moisture, ash and carbohydrate contents of annular seabream were found as 17.78%, 2.28%, 76.96%, 1.80% and 1.19%, respectively (Figure 2). The energy value of the annular seabream was calculated as 96.35 kcal/100g.

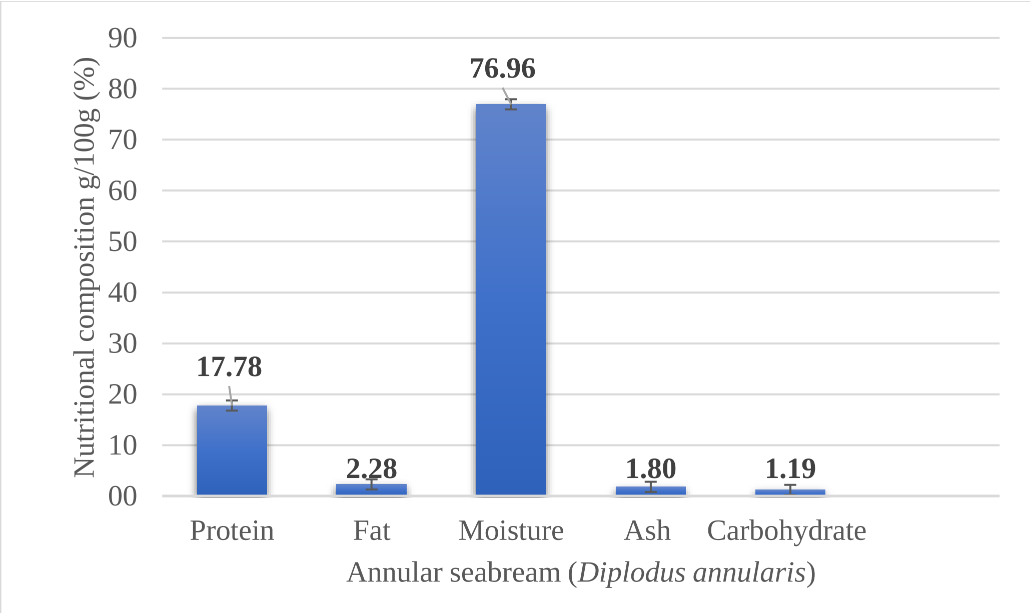


Figure 2. *Nutritional composition of the annular seabream (Diplodus annularis) (g 100g/wet weight (ww))*

No study has been found in literature investigating the nutritional composition of annular seabream inhabiting the Black Sea. Moreover, Smichi et al. [41] pointed out that there is no published data on the nutritional quality and composition of annular seabream, although it has a wide distribution in the Mediterranean. However, Ketata-Khitouni et al. [17] examined the protein, fat, moisture and ash amounts of samples taken from different body parts of the male and female annular sea breams, separately. In another study, the chemical composition of sea bream was investigated seasonally. When compared with the sampling conducted by the researchers in May, the moisture and protein % values of the data obtained were found to be close to our study, while the fat and ash values were higher than those found in our study [18]. Smichi et al. [41] calculated the protein, fat and moisture values of annular seabream on dry weight. Accordingly, the % moisture value of annular sea bream was reported as 18.67% based on dry matter. Ozogul et al. [21] reported the fat value of annular sea bream as 2.52%. In another study, Passi [19] reported this value as 2.32%. It can be said that these fat values are quite close to the values found in our study.

Amino Acids

The amino acid composition of annular seabream is shown in Table 2. Seafood is one of the most aromatic foods. It has a particularly delicious and different aroma compared to other foods, especially because it contains a high proportion of glutamic acid [42]. The first three amino acids most abundant in annular seabream were glutamic acid, aspartic acid and alanine, respectively. However, the least abundant ones were cystine, tyrosine and histidine, respectively. While the total amount of essential amino acid was 147.45 mg/g, the total amount of non-essential amino acid was measured as 462.74 mg/g. The ratio of essential/non-essential amino acids (E/NE) was observed to be 0.32 in the annular seabream. The amount of total sweet amino acids was found to be higher than the total bitter amino acids.

Table 2. *Amino acid composition of annular seabream (Diplodus annularis)*

Amino acids (mg/g / ww)	Annular seabream (*Diplodus annularis*)
Alanine	56.39±2.33
Leucine*	18.24±1.57
Aspartic acid	100.57±1.94
Arginine	13.96±1.20
Cystine	1.48±0.14
Glutamine	15.16±1.07
Glutamic acid	225.40±25.93
Histidine*	8.07±1.30
Isoleucine*	18.12±1.87
Lysine*	15.10±1.33
Methionine	9.41±2.21
Phenylalanine*	8.83±0.23
Proline	23.27±2.11
Serine	33.42±0.65
Threonine	43.32±2.34
Tyrosine	7.05±0.22
Valine*	12.42±0.42
Total amino acids	**610.19±39.92**
Total essential amino acids (E)*	**147.45±3.65**
Total non-essential amino acids (NE)	**462.74±32.02**
E/NE	**0.32±0.00**
Sweet amino acids	**315.22±27.52**
Bitter amino acids	**55.35±5.73**

All samples were analyzed in duplicate. Values are shown as mean ± standard error

No study has been found in literature investigating the amino acid composition of annular seabream (*Diplodus annularis*). However, in recent years, protein hydrolyzate studies have been carried out on this species [43, 44]. Kouroupakis et al. [45] studied the amino acid composition of the species *Diplodus* *sargus*. When the data obtained in the aforementioned study were compared with the present study, it was observed that glutamic acid was the highest amino acid in both studies. Similarly, total essential amino acids were lower than total non-essential amino acids in both studies.

Fatty Acids

The fatty acid composition of annular seabream is shown in Table 3. The ΣSFA amount was found to be 34.85%. While the highest amount of saturated fatty acid was found as palmitic acid (15.86%), the lowest amount of saturated fatty acid was tridecanoic acid (0.02%). The amount of ΣMUFA was 27.36%. Oleic acid was the highest monounsaturated fatty acid with 19.42%, whereas erucic acid was the lowest with 0.11%. The amount of ΣPUFA was measured as 37.74%. The highest amounts of polyunsaturated fatty acids were detected as linoleic acid (8.78%) and docosahexaenoic acid (8.76%), respectively. These fatty acid values were found to be close to each other. However, the lowest amount of polyunsaturated fatty acid was docosadienoic acid with a value of 0.16%. The amounts of ω3 and ω6 were 19.00% and 17.36%, respectively. Accordingly, the ω3/ω6 ratio was calculated as 1.09%. Ketata-Khitouni et al. [18] examined the seasonal fatty acid profile of annular sea bream. The total amount of SFA, MUFA and PUFA in spring was reported as 42.43%, 35.32% and 22.24% in males, and 41.28%, 37.17% and 21.54% in females, respectively. In another study, the ΣSFA, ΣMUFA and ΣPUFA contents were reported as 28.73%, 54.66 %and 16.6%, respectively [41]. PUFA values in our study were found to be higher than those found in both of these aforementioned studies. However, SFA and MUFA values are different from each other. Another research was carried out on fish caught off the coasts of Tunisia. There are many factors (such as catch location, catch time, nutritional status, age, and gender) that affect the nutritional composition of fish in general. Passi et al. [19] and Bouhlel et al. [20] reported that the amounts of palmitic acid, oleic acid and docosahexanoic acid in annular seabream were higher than the other fatty acids detected. These results are similar to our study.

Lipid Quality Indices (LQI)

Lipid Quality Indices (LQI) of annular seabream are shown in Table 4. In the simplest terms, the Atherogenicity Index (AI) shows the relationship between total major saturated fatty acids and total major unsaturated fatty acids. A low AI value is desirable to prevent micro and macro coronary diseases. The Thrombogenicity Index (TI) indicates a tendency to form clots in blood vessels. It is defined as the relationship between pro-thrombogenetic (saturated) and anti-thrombogenetic fatty acids (MUFAs, PUFAs – n6 and PUFAs – n3) [30]. It has been reported that AI and TI values should be lower than 1 to prevent heart diseases [30, 46]. The AI and TI values obtained in our study were within the recommended values.

Table 3. *Fatty acid composition of annular seabream (Diplodus annularis) (g 100g/wet weight(ww))*

Fatty acid (%)		**Annular seabream**
C12:0	Lauric acid	0.12±0.00
C13:0	Tridecanoic acid	0.02±0.00
C14:0	Myristic acid	3.44±0.06
C15:0	Penta decanoic acid	0.91±0.04
C16.0	Palmitic acid	15.86±0.24
C17:0	Heptadecanoic acid	1.33±0.00
C18:0	Stearic acid	8.65±0.11
C20:0	Arachidic acid	0.84±0.02
C21:0	Heneicosanoic acid	0.04±0.01
C22:0	Behenic acid	0.57±0.03
C23:0	Tricosanoic acid	0.24±0.00
C24:0	Lignocerik acid	2.85±0.07
ΣSFA		**34.85±0.32**
C14:1	Myristoleic acid	0.40±0.01
C15:1 cis-10	Pentadecenoic acid	0.22±0.00
C16.1	Palimiteloic acid	1.57±0.04
C17:1 cis-10	Heptadecanoic acid	0.87±0.01
C18:1n9c	Oleic acid	19.42±0.02
C18:1n9t	Elaidic acid	1.96±0.09
C20:1 cis-11	Eicosenoic acid	1.88±0.01
C22:1n9	Erucic acid	0.11±0.01
C24:1	Nervonic acid	0.95±0.07
ΣMUFA		**27.36±0.07**
C18:2n6t	Linoleadic acid	0.56±0.04
C18:2n6c	Linoleic acid	8.78±0.03
C18:3n3	a-Linolenic acid	2.99±0.03
C18:3n6	g-Linolenic acid	0.33±0.01
C20:2 cis 11,14	Eicosadienoic acid	1.23±0.02
C20:3n3 cis 11,14,17	Eicosatrienoic acid	1.54±0.01
C22:2 cis 13,16	Docosadienoic acid	0.16±0.01
C20:5n3 cis 5,8,11,14,17	Eicosapentanoic acid	5.71±0.02
C22:6n3 cis-4,10,13,16,19	Docosahexanoic acid	8.76±0.15
C20:4n6	Arachidonic acid	6.46±0.10
C20:3n6 cis-_8_11_14	Eicosatrienoic acid	1.23±0.02
ΣPUFA		**37.74±0.36**
ω3		**19.00±0.22**
ω6		**17.36±0.13**
Unidentified		**0.06±0.03**
ω3/ω6		**1.09±0.00**

All samples were analyzed in triplicate. Values are shown as mean ± standard error

Table 4. *Lipid Quality Indices (LQI) of annular seabream (Diplodus annularis)*

Indices	**Annular seabream**	**References**
AI (Atherogenicity Index)	0.47±0.00	Ulbricht and Southgate [30]
TI (Thrombogenicity Index)	0.35±0.00	Ulbricht and Southgate [30]
PI (Polyene Index)	0.91±0.02	Lubis and Buckle [31]
H/H (Hypocholesterolemic/hypercholesterolemic ratio)	2.83±0.05	Santos-Silva et al. [32]
FLQ (Fish Lipid Quality)	14.48±0.17	Abrami et al. [33]

Values are shown as mean ± standard error

The Polyene Index (PI) was used as a measure of PUFA damage [31]. In our study, the PI value was calculated as 0.91. An increase in the amount of cholesterol in the blood is called hypercholesterolemia, and a decrease in it is called hypocholesterolemia [47]. In the current study, the Hypocholesterolemic/Hypercholesterolemic (H/H) ratio was 2.83. This ratio should be high in terms of healthy nutrition. Based on the total lipids, fish lipid quality (FLQ) demonstrates the percentage of the main HUFA-n3 (EPA and DHA) in total fatty acids of the muscle. The higher the value of this index, the higher the quality of the dietary lipid source [33]. In the current study, the FLQ value of annular seabream was found as 14.48.

Minerals and Heavy Metals

Mineral and heavy metal contents of annular seabream are shown in Table 5 and Table 6. The mineral content of muscle foods such as seafood is relatively variable and depends on several factors. Some of these factors can be considered as nutrition, species, breed, sex, age at slaughter, muscle types, physiological status, production system, processing methods and method of analysing the mineral content of the meat samples [48 – 50]. In the present study, the concentrations of K, P, Na, Ca and Mg minerals in the annular sea bream were in the order of K>P> Na>Ca>Mg. The potassium (K) and phosphorus (P) content of the annular seabream were 6228.02 mg/kg and 3726.48 mg/kg, respectively. The rarest minerals in the annular seabream were Cs, Mo, Gd, Ni and Sb, respectively. Ketata-Khitouni et al. [18] reported that annular sea bream was rich in potassium (K) and calcium (Ca) in all seasons. In another study, Smichi et al. [41] reported the main minerals in annular seabream as sodium (Na), calcium (Ca) and magnesium (Mg). However, in the present study, P was found to be one of the most abundant main elements, along with K, Na, Ca and Mg. In the current study, Fe, Zn, Cu and Ni values were measured as 8.59 mg/kg, 7.89 mg/kg, 0.30 mg/kg and 0.02 mg/kg, respectively. These values were reported as 12.20 mg/kg, 135.77 mg/kg, 0.11 mg/kg and 0.66 mg/kg in another work [51]. It was observed that the findings of both studies were different from each other. Bat [52] reported the Fe, Zn and Cu values of annular sea bream in the Black Sea as 19.3 mg/kg, 10.2 mg/kg and 0.17 mg/kg, respectively. Accordingly, while the Fe and Zn values in our study were lower than the values of Bat [52], the Cu value was found to be higher.

Table 5. *Mineral contents of annular seabream (Diplodus annularis)* (mg kg^{-1}, ww)

Mineral elements	Annular seabream (mg kg-1, ww)
Na	900.88±0.59
Mg	454.82±2.20
K	6228.02±10.03
P	3726.48±20.05
Ca	584.83±0.15
Mn	0.22±0.00
Se	0.50±0.00
Si	10.21±0.16
Fe	8.59±0.04
Zn	7.89±0.08
Al	4.09±0.03
Rb	1.13±0.00
Sr	1.95±0.01
Cr	0.83±0.00
As	0.71±0.01
Ti	0.59±0.02
Be	0.57±0.00
Cu	0.30±0.00
Li	0.05±0.00
Ba	0.08±0.00
Gd	0.02±0.00
Ni	0.02±0.00
Sb	0.02±0.00
Cs	0.01±0.00
Mo	0.01±0.00
Hg	0.15±0.00
Pb	0.02±0.00
Cd	0.01±0.00

All samples were analyzed in triplicate,
Values are shown as mean ± standard error, ww. wet weight

In a study conducted with the Black Sea annular seabream, Cd, Hg and Pb values were reported as 0.01 mg/kg, 0.02 mg/kg and 0.12 mg/kg, respectively [52]. Cd values of the current study were similar to those given by Bat [52]. However, in the current study, Hg values were found to be higher, while Pb values were lower. Ben Salem and Ayadi [51] reported Pb and Cd values in annular sea bream as 0.17 mg/kg and 0.76 mg/kg, respectively. The same researchers reported the Hg value as 0.20 mg/kg [53]. The European Commission has set maximum levels for heavy metals in fish muscle. The legal limits for Hg, Pb, and Cd are 500, 300, and 50 µg/kg w.w., respectively [54]. The values of Hg, Pb, and Cd in the muscles of fish in the current study were below the maximum limits set by Commission Regulation (EC) [54].

Public Health Risk Analysis

Metal Pollution Index (MPI)

The MPI value of the annular seabream was calculated as 0.23. Gencer and Kocatepe [55] reported that high heavy metal contamination in the samples increased the MPI value. Haseeb-ur-Rehman et al. [56]

reported in a study that the MPI values of various seafood products varied between 0.62 and 2.78. Compared to these values, the metal pollution of annular seabream can be considered as quite low.

Estimated Daily Intake (EDI), Estimated Weekly Intake (EWI)

The EDIs and EWIs of metals through the consumption of annular seabream muscles are shown in Table 6. EWI values of Cd, Hg and Pb were compared with the Provisional Tolerable Weekly Intake (PTWI) to assess public health risks. The PTWI values were assumed to be 0.007 mg for Cd, 0.004 mg for Hg [38] and 0.025 mg for Pb [57].

Table 6. *EDIs and EWIs of metals via consumptions of annular seabream muscles*

Elements	PTWI*	PTWI**	EDI	EWI
Cd	0.007	0.49	2.86E-06	2.00E-05
Pb	0.025	1.75	5.71E-06	4.00E-05
Hg	0.004	0.28	4.29E-05	3.00E-04

* Internationally recommended PTWI safe levels for the studied metals.
Provisional tolerable weekly intake in mg/week/kg body weight [38]
**PTWI for a 70 kg adult (mg/week/kg body weight)
EDI Estimated Daily Intake (mg/day body weight)
EWI Estimated Weekly Intake (mg/week body weight)

The Estimated Weekly Intake (EWI) values for metals are based on an adult's weekly consumption. Annular seabream's EDI and EWI values were found to be below the recommended standard reference values.

Conclusion

In the current study, meat yield, nutritional composition, amino acids, fatty acids, lipid quality indices, and mineral and heavy metal contents of annular seabream caught in the Black Sea were investigated. Accordingly, annular seabream was observed to have high protein and low fat content. The most abundant amino acids in annular sea bream meat were found as glutamic acid, aspartic acid and alanine. In addition, polyunsaturated fatty acids (PUFAs) were the predominant fatty acids. AI and TI values were found to be less than 1. One can say that annular seabream is rich in potassium, phosphorus and sodium minerals. Additionally, the amounts of Hg, Pb and Cd were found to be within the permitted limits for consumption. As a result, annular seabream *(Diplodus annularis)* can be an alternative species that can be consumed in healthy diets since it contains high protein, low fat, rich essential amino acids, high quality fatty acids and important minerals. However, considering the annually declining stocks, future studies should focus on the reasons for the low population density of this valuable fish.

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