

## Investigation and Comparison of the Suitability and Usability of Fatty Acids and the Lipid Quality Index Parameters in Waste Skin and Bones of Some Cultured Fish

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### Abstracts

In this study, the suitability and usability of fish skin and bones generated as waste in terms of lipids and fatty acids were investigated. In this paper, the cultured *Sparus aurata* (Sea bream), *Dicentrarchus labrax* (Sea Bass) and *Oncorhynchus mykiss* (Rainbow trout) fish were examined. Within the scope of the study, the fatty acids and lipid quality index parameters in the skin and bones of the cultured fish were calculated. The lipid quality index was examined for skin and bone of cultured fish using the Atherogenicity Index (AI), Thrombogenicity Index (TI), Flesh-lipid quality (FLQ), Hypocholesterolemic/hypercholesterolemic ratio (h/H), Health-promoting index (HPI), Unsaturation Index (UI), The polyene index (PI), Hypercholesterolaemic fatty acids (OFA), and Desirable fatty acids (DFA). Among these indices, the AI values were obtained between 0.18 and 0.24 in all fish bone and skins. The TI values also obtained between 0.71 and 0.98 for all bone and skins. Among the important fatty acids, EPA was calculated between 0,34 and 2.22g/100g in skins and 0.29 and 0.87g/100g in bones. According to our results, it has been determined that the fatty acids levels of  $\omega$ -3,  $\omega$ -6, and Linoleic acid (LA) in fish skin and bones based on 100g of dry weight composition (DWC) are higher than the daily consumption levels recommended by The Australian National Health and Medical Research Council (NHMRC). In conclusion, it can be said that the skin and bones of cultured fish can be used for dietary consumption purposes.

**Keywords:** Fatty Acid; Lipid Quality Index; EPA; Atherogenicity ve Thrombogenicity Index.

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## 1. Introduction

In today's fast-paced world where food consumption is rapidly increasing, seafood, particularly fish, which is rich in high-quality proteins, vitamins, minerals, and polyunsaturated fatty acids (PUFAs) play an important role as a part of a balanced diet (Ehsani and Jasour, 2012; Dağtekin *et al.*, 2018). Fatty acids are organic compounds that contain a carboxyl (-COOH) group and single or double bonds in their structure. Fatty acids with single bonds are called saturated fatty acids, while those with double bonds are called unsaturated fatty acids (Chen and Liu, 2020). Fatty acids play many roles in our body. They contribute to the formation of hormones and the preservation of cell structure. They also can be used to store energy in the body. It is known that fatty acids are distributed throughout living cells and provide energy for metabolism in the body. Additionally, it is known that fatty acids play a critical role in human metabolism, health, and disease (Chen and Liu, 2020). Recent studies, particularly epidemiological, clinical, and nutritional research have found that the consumption of foods rich in polyunsaturated fatty acids (PUFAs) has positive effects on cardiovascular diseases, diabetes, obesity, arthritis, asthma, depression, hyperactivity, and certain types of cancer (Ruxton *et al.*, 2004; Gogus and Smith, 2010; Dağtekin *et al.*, 2018).

Fatty acids obtained from many different food sources affect human health. Therefore, it is important to determine the fatty acid content in foods, food supplements, and herbal medicines to determine their nutritional and/or medicinal value (Chen and Liu, 2020). Essential fatty acids are important for human health and it is known that the human body lacks the enzymes necessary to produce the two basic fatty acids EPA and DHA. Therefore, these fatty acids must be obtained from external sources through diet. In particular, marine fish are important food sources because they are rich in docosahexaenoic acid (DHA) and eicosapentaenoic acid (EPA) (Öksüz *et al.*, 2011; Ghaeni and Ghahfarokhi, 2013). In recent years, there has been a focus on studying the lipid composition of fish and fish products that are rich in n-3 fatty acids. Fatty acids in health-beneficial foods affect the quality and quantity of the nutritional value and foods that contain high-quality fatty acids are also gaining commercial interest. For example, fatty acids such as omega-3 and omega-6, which are rich in these fatty acids, have been shown by some studies to be beneficial for heart health (Garaffo *et al.*, 2011). It is stated that polyunsaturated fatty acids (PUFAs) found in fish have significant effects on human health by reducing the risk of stroke, lowering serum triglyceride levels, decreasing blood pressure and insulin resistance, and regulating glucose metabolism (Duo *et al.*, 2003; Garaffo *et al.*, 2011).

## 2. Materials and Methods

### 2.1. Preparation of fish skins

In this study, skin and bone of cultured *Sparus aurata* (Sea bream), *Dicentrarchus labrax* (Sea Bass), and *Oncorhynchus mykiss* (Rainbow trout) were obtained from Canakkale fish market. The fish to be used for the production of gelatin were firstly subjected to the filleting process without damaging their bones and skin, and then the skin and bones were carefully removed from the separated fillets.

## 2.2. Crude oil extraction

In order to determine the fatty acid composition of crayfish, the samples were homogenized and dried at 105 °C until they reached a constant weight in a drying oven. The extraction of crude fat was performed three times on the dry tissue. The commonly used Bligh and Dyer (1959) method was used for fat analysis. In summary, the samples were treated with methanol/chloroform. The homogenate was washed with methanol-chloroform and filtered through filter paper into a round-bottomed flask. The filtrate was evaporated using a rotary evaporator (IKA RV10 basic) in a water bath at 60°C. After the separation of fat in the round-bottomed flask, the flask was removed from the device and kept in a drying oven at 65°C (Nüve FN500), then transferred to a desiccator and cooled, and finally weighed for the final determination.

## 2.3. Esterification of fatty acids

The fatty acid analysis was performed according to AOAC (1995). The crude fats of the samples were used. In summary, the crude fat samples were treated with methanolic NaOH to esterify the crude fat. Then, it was saponified by boiling in a water bath. After BF<sub>3</sub> reagent was poured over the cooler, it was heated and heptane was added. After that, it was cooled again without boiling and treated with saturated NaCl to form a phase. The heptane phase was then transferred to a test tube and transferred to a glass vial. Next, it was injected into gas chromatography (GC) to determine the fatty acid composition.

## 2.4. Determination of fatty acid contents by Gas Chromatography (GC)

Shimadzu GC (Gas Chromatography) was used to determine the fatty acid components. The system consists of FID detector (Flame Ionization Detector), a gas chromatograph (Shimadzu, GC 2014, Japan) and autoinjector (AOC-20i, Shimadzu, Japan). The device is controlled by GC solution software (Version 2.41.00 su\_1). FAME WAX (polyethylene glycol, 30 meter 0.25 mm I.D. 0.2 µm, GC Columns Restek) was used as the chromatographic column. The data were obtained by calculating the methyl esters of fatty acids as a percentage of total fatty acids. Supelco 37 Component FAMES Mix standard (Sigma-Aldrich) was used to determine the fatty acid peaks. The GC spectrum of fatty acids and GC operating conditions are given in Figure 1.

The percentage of fatty acid composition obtained from GC was used to calculate the amounts of fatty acids in the samples in mg/100 g portion as edible fats according to Weihrauch *et al.* (1977) using the fatty acid conversion factor.

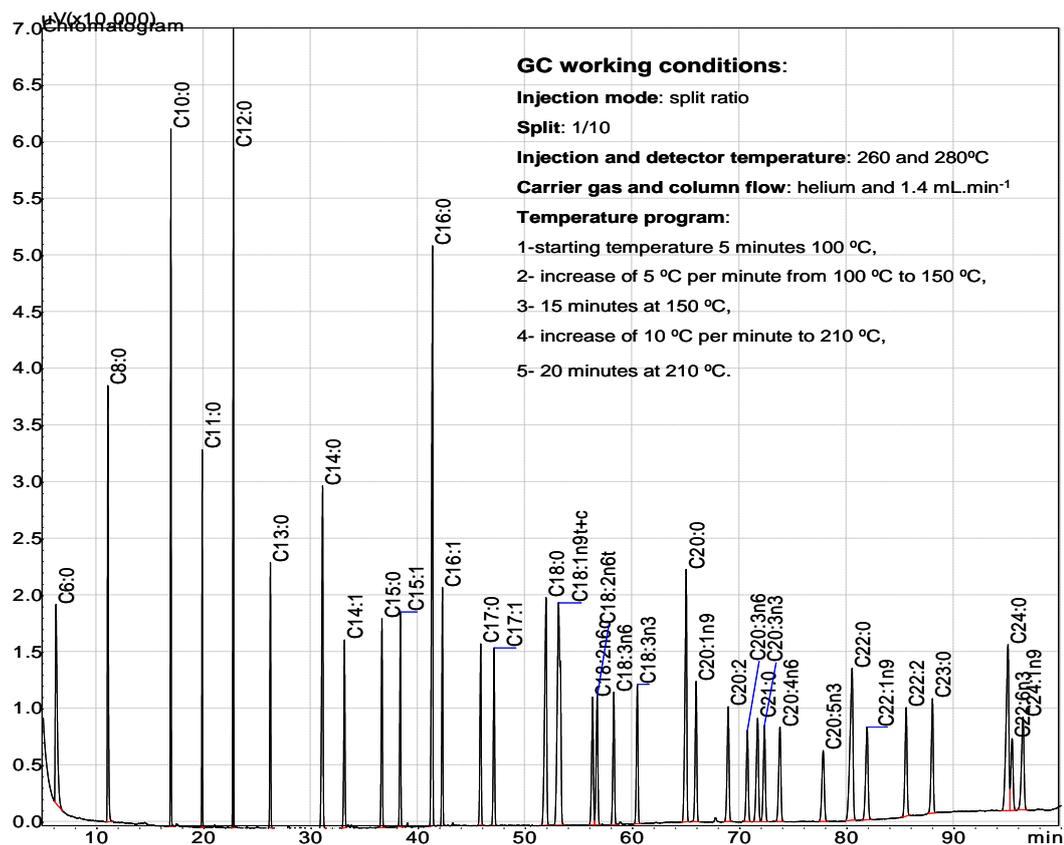


Figure 1. GC spectrum of fatty acids (original) and GC working conditions

## 2.5. Calculation of the Lipid Quality Indexes (LQI)

The lipid quality index was calculated using the fatty acid profile obtained from gas chromatography analyses and results. In this context, nine different calculation methods were used for the lipid quality index. LQI calculations were obtained using percent fatty acids results and the calculated LQI results are percent values (%). These calculation methods are as follows:

- 1- Atherogenicity Index (AI):  $[C12:0 + (4 \times C14:0) + C16:0] / (n\text{-}3\text{PUFA} + n\text{-}6\text{PUFA} + \text{MUFA})$  (Ulbricht and Southgate, 1991; Garaffo *et al.*, 2011; Luczynska and Paszczyk, 2019)
- 2- Thrombogenicity Index (TI):  $[C14:0 + C16:0 + C18:0] / [(0.5 \times C18:1) + (0.5 \times \text{sum of other MUFA}) + (0.5 \times n\text{-}6\text{PUFA}) + (3 \times n\text{-}3\text{PUFA}) + n\text{-}3\text{PUFA}/n\text{-}6\text{PUFA}]$  (Ulbricht and Southgate, 1991; Garaffo *et al.*, 2011; Łuczynska and Paszczyk, 2019)
- 3- Flesh-lipid quality (FLQ):  $100 \times (\text{EPA} + \text{DHA}) / \text{total fatty acids}$  (Abrami *et al.*, 1992; Senso *et al.*, 2007; Łuczynska and Paszczyk, 2019)
- 4- Hypercholesterolaemic fatty acids (OFA):  $C12:0 + C14:0 + C16:0$  (Łuczynska and Paszczyk, 2019)
- 5- Desirable fatty acids (DFA):  $C18:0 + \text{UNSAT}$  (Costa *et al.*, 2008; Silva *et al.* 2019; Łuczynska and Paszczyk, 2019)

- 6- Hypocholesterolemic/hypercholesterolemic ratio (h/H):  $h/H = [(C18:1 + C18:2 + C18:3 + C20:3 + C20:4 + C20:5 + C22:4 + C22:5 + C22:6) / (C14:0 + C16:0)]$  (Santos-Silva *et al.*, 2002, Dagtekin *et al.*, 2018)
- 7- Health-promoting index (HPI):  $UNSAT / [C12:0 + (C14:0 \times 4) + C16:0]$  (Chen and Liu, 2020)
- 8- Unsaturation Index (UI):  $1 * (\% \text{ monoenoics}) + 2 * (\% \text{ dienoics}) + 3 * (\% \text{ trienoics}) + 4 * (\% \text{ tetraenoics}) + 5 * (\% \text{ pentaenoics}) + 6 * (\% \text{ hexaenoics})$  (Logue *et al.*, 2000; Chen and Liu, 2020)
- 9- The polyene index (PI):  $(C20:5 + C22:6) / C16:0$  (Lubis and Buckle, 1990; Küçükgülmez *et al.*, 2018).

### 3. Results and Discussion

In recent years, the quality and diversity of the components that make up the feed and food sources have gained increasing importance due to the human population. As a result, research has focused on the development of food sources. It is known that fishes in seafood provide an important source of protein, mineral, oil and amino acid in nutrition (Kale, 2020). One of the important nutrients in food sources is fats and fatty acids. The diversity and amount of fatty acids that make up the structure of fat are very important for the development and continuity of human metabolism. In particular, fatty acids such as  $\omega$ -3 (omega-3), EPA (Eicosapentaenoic Acid), and DHA (Cis-4,7,10,13,16,19-Docosahexaenoic Acid) are well known for their importance. In this regard, the quality and quantity of fat and fatty acids in a food source at the consumption stage are of great importance. It is stated that the temporal and spatial distribution of fish is affected by biotic and environmental factors (Thrush *et al.*, 2001; Altin *et al.*, 2020). For example, mercury which is one of the toxic elements can accumulate in the environment and living metabolism (Astani *et al.*, 2011; Shahid *et al.*, 2020; Parang *et al.*, 2020). On the other hand, aquaculture, being performed in a specific sea area, is carried out under controlled conditions.

#### 3.1. Fatty acid compositions

In this study, the fat compositions and quality index parameters in cultured fish skin and bones were examined. The fatty acid compositions of skin and bones are given in Table 1. C4:0, C6:0, C8:0, C10:0, C11:0, C12:0, C13:0, C14:0 and C15:0 fatty acids were not determined that it was below the chromatography analysis measurement limits in all samples. Therefore, these fatty acids were not included in the table. Table 1 shows the results and evaluations of fatty acids using two different calculation methods. Table 1 represents the percentage values in all fatty acids (%) and gram amount in 100 g dry weight compositions of the fatty acid components in the crude fat. The dry weight composition (g/100 g) is named as dry sample compositions (DWC). This study uses both methods to facilitate comparison with other studies. The DWC were calculated using fatty acid conversion factor (Weihrach *et al.*, 1977). The amounts of fat in the skin and bones of cultured fish are given in Table 1. The highest fat content was found in plaice skin at 62.2%. The fat content in other samples ranged from 29% to 33%. The approximate percentage of total unsaturated fatty acids for all samples is 55%. The amount of unsaturation was calculated as 15 to 33 g/100 g as DWC. The ratio of

unsaturation to saturation (UNSAT/SAT) is between 1 and 1.2 in percentage calculation, while it is below 1 in DWC calculation. This shows that different results can be obtained when fatty acid ratios are calculated based on the amount of crude oil in the dry sample (DWC). It can be inferred that these calculation methods can be helpful in obtaining comparable results.

In general, when Table 1 is evaluated, it is seen that the highest and dominant fatty acid C18 has an average of 25%, which constitutes approximately  $\frac{1}{4}$  of the total fatty acids. The C18:0 fatty acid found in salmon and sea bream bones is more than in their skin, while in sea bass it is the opposite, with more C18 fatty acid found in the skin samples. Generally, C18:0 fatty acid ranges from 25% to 32% and it was found to be higher in the skin of both cultured fish than in their bones. On the other hand, when the DWC amounts of C18:0 fatty acid were examined, they varied between 7 and 15 g/100 g. When the calculation is done by DWC method which includes the amount of oil in the dry sample, it is calculated that there is more C18:0 fatty acid in the skin of salmon fish. Therefore, DWC calculation can provide different results compared to percentage calculation (%). C (Cis)+T (Trans) in Table1 represents cis and trans fatty acids. C18:1n9C+T is difficult to differentiate chromatographically because they are different isomers of the same molecule. For this reason, C18:1n9 is represented by two isomers in chromatography. C18:1n9C+T is seen as the second highest fatty acid. It was calculated between 11 and 16 % in percentage and between 3 and 10 g/100 g in the dry weight composition (DWC).

$\omega$ -3,  $\omega$ -6, DHA/EPA,  $\omega$ -3/ $\omega$ -6, PUFA/SAFA, and UNS/SAFA are values used to measure the nutritional value of fats (Prato *et al.*, 2018).  $\omega$ -3 fatty acids are known to be important for human health and are a important dietary source (Howe *et al.* 2006). Eicosapentaenoic acid (EPA; 20:5,  $\omega$ -3) and docosahexaenoic acid (DHA; 22:6, n-3) are easily usable in metabolism and play a vital role in some metabolic activities (Babu *et al.* 2012). In this study, EPA was found to range from 1 to 3.75% and DHA from 3 to 6%. DWC for EPA was calculated to be between 0.3-2 g/100 g, and DHA between 1-2.75 g/100 g. Total  $\omega$ -3 fatty acids were determined to be between 5-9% and DWC between 1.5 and 5 g/100 g. Total  $\omega$ -6 fatty acids were determined to be between 31-43% and DWC between 9 and 18 g/100 g. In scientific studies, the higher the total omega-6/3 ratio, the higher the risk of the pathogenesis of many diseases, cardiovascular disease, cancer, inflammatory and autoimmune diseases (Simopoulos, 2008; Stratev *et al.*, 2017). For these reasons, such calculations are important. It is recommended that the total omega-6/3 ratio should not exceed 4 (Stratev *et al.*, 2017). It is stated that the type of fat is more important than the total amount of fat in measuring the risk of cardiovascular disease (Stratev *et al.*, 2017). The n-3/n-6 ratio is a good index for comparing the relative nutritional value of fish oil (Küçükgülmez *et al.*, 2018). A high n-3/n-6 ratio is important in reducing the risks of coronary heart disease, plasma lipid levels, and cancer (Kinsella *et al.* 1990; Küçükgülmez *et al.*, 2018). In our study, the total omega-6/3 ratio for female individuals varies between 3-7%.

Table 1. Percentage (%) and calculated gram amount in the dry weight composition (DWC, g/100 g) of fatty acids on cultured fish skin and bones

	OM Skin		OM Bone		SP Skin		SP Bone		DL Skin		DL Bone	
	%	DWC										
C16:0	15.62	4.66	11.33	3.22	14.58	8.64	13.93	4.41	11.62	3.42	15.43	4.52
C16:1	4.83	1.44	2.21	0.63	5.51	3.26	5.57	1.77	2.18	0.64	3.97	1.16
C17:0	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C17:1	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C18:0	25.37	7.57	32.86	9.33	27.67	16.39	30.57	9.69	32.05	9.42	27.41	8.03
C18:1n9C+T (ω-9)	16.90	5.04	11.40	3.24	13.20	7.82	11.61	3.68	10.39	3.05	15.44	4.52
C18:2n6c (ω-6), LA	15.75	4.70	26.65	7.57	16.45	9.74	16.40	5.20	26.83	7.89	16.33	4.79
C18:2n6t (ω-6)	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C18:3n6 (ω-6)	3.14	0.94	3.56	1.01	3.28	1.94	3.37	1.07	3.69	1.08	3.20	0.94
C18:3n3 (ω-3), ALA	0.50	0.15	0.58	0.16	0.53	0.31	0.53	0.17	0.95	0.28	0.40	0.12
C20:0	0.24	0.07	0.11	0.03	0.27	0.16	0.24	0.08	0.19	0.06	0.22	0.06
C20:1n9 (ω-9)	3.19	0.95	2.08	0.59	2.60	1.54	2.73	0.87	1.91	0.56	3.65	1.07
C20:2	0.96	0.29	1.26	0.36	1.06	0.63	0.94	0.30	1.23	0.36	1.16	0.34
C20:3n6 (ω-6)	0.21	0.06	0.84	0.24	0.27	0.16	0.30	0.10	0.76	0.22	0.21	0.06
C21:0	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
C20:3n3 (ω-3)	0.64	0.19	0.58	0.16	0.45	0.27	0.43	0.14	0.59	0.17	0.53	0.16
C20:4n6 (ω-6)	0.22	0.07	0.15	0.04	0.37	0.22	0.33	0.10	0.16	0.05	0.24	0.07
C20:5n3 (ω-3), EPA	3.75	1.12	0.99	0.28	2.85	1.69	2.75	0.87	1.15	0.34	2.85	0.84
C22:0	0.06	0.02	0.53	0.15	0.52	0.31	0.04	0.01	0.20	0.06	0.47	0.14
C22:1n9 (ω-9)	1.15	0.34	0.33	0.09	0.85	0.50	1.46	0.46	0.78	0.23	0.93	0.27
C22:2	0.08	0.02	0.05	0.01	0.18	0.11	0.16	0.05	0.14	0.04	0.23	0.07
C23:0	0.11	0.03	0.11	0.03	0.13	0.08	0.18	0.06	0.16	0.05	0.12	0.04
C22:6n3 (ω-3), DHA	4.64	1.38	3.20	0.91	6.32	3.74	5.40	1.71	3.74	1.10	4.72	1.38
C24:1n9 (ω-9)	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
UNSAT	55.93	16.68	53.87	15.30	53.92	31.93	51.98	16.47	54.50	16.02	53.88	15.79
SAT	44.07	13.15	46.13	13.10	46.08	27.29	48.02	15.22	45.50	13.38	46.12	13.51
Σ MUFA	26.07	7.78	16.02	4.55	22.16	13.12	21.37	6.77	15.26	4.49	23.99	7.03
Σ PUFA	29.89	8.92	37.86	10.75	31.76	18.81	30.61	9.70	39.24	11.54	29.87	8.75
UNSAT/SAT	1.27	0.38	1.17	0.33	1.17	0.69	1.08	0.34	1.20	0.35	1.17	0.34
Σ PUFA/SAT	0.68	0.20	0.82	0.23	0.69	0.41	0.64	0.20	0.86	0.25	0.65	0.19
DHA/EPA	1.24	0.37	3.23	0.92	2.22	1.31	1.96	0.62	3.25	0.96	1.66	0.49
Total ω-3	9.53	2.84	5.35	1.52	10.15	6.01	9.11	2.89	6.43	1.89	8.50	2.49
Total ω-6	32.03	9.56	41.77	11.86	34.60	20.49	33.19	10.52	43.18	12.70	31.95	9.36
Total ω-9	21.24	6.34	13.81	3.92	16.65	9.86	15.80	5.01	13.08	3.85	20.02	5.87
ω-6/ ω-3	3.36	3.36	7.81	7.81	3.41	3.41	3.64	3.64	6.72	6.72	3.76	3.76
Crude Oil (g/100g)	31.45		29.95		62.20		33.40		31.00		30.90	

OM: and *Oncorhynchus mykiss* (Rainbow trout), SP: *Sparus aurata* (Sea bream), DL: *Dicentrarchus labrax* (Sea Bass)

### 3.2. The lipid quality indexes (LQI)

In this study, LQI values of the oil are calculated using the percentage results (%) of fatty acids in the samples and LQI values are percentage results (%). The LQI values in our study are given in Table 2. The lipid quality index is based on important calculations that are commonly used in fatty acid determination studies by many researchers. The atherogenicity index (AI) shows the

relationship between the total of the main saturated fatty acids and the total of the main unsaturated classes. Saturated (SAT) fatty acids that contain C12:0, C14:0, and C16:0, except for C18:0, are considered proatherogenic (they facilitate the adhesion of lipids to circulatory and immunological system cells) (González-Félix *et al.*, 2016; Monteiro *et al.*, 2018; Omri *et al.*, 2019; Chen and Liu, 2020). Lipids support the adhesion of immune and circulatory system cells (Proatherogenic) and are anti-atherogenic, inhibiting platelet aggregation and reducing levels of esterified fatty acid, cholesterol, and phospholipids, preventing the occurrence of micro and macro coronary diseases (anti-atherogenic) (Ghaeni and Ghahfarokhi, 2013). AI is commonly used for foods such as seaweed, cereals, meat, fish, and dairy products (Chen and Liu, 2020). AI values were determined to range between 0.18-0.24. TI indicates the tendency to form thrombi in blood vessels and provides the contribution of different FA's showing the relationship between pro-thrombogenic FA's (C12:0, C14:0, and C16:0). The thrombogenicity index (TI) characterizes the thrombogenic potential of fatty acids and indicates the tendency to form thrombi in blood vessels. It is defined as the relationship between pro-thrombogenic (saturated) and anti-thrombogenic fatty acids (MU-FAs, PUFAs - n-6, and PUFAs - n3) (Garaffo *et al.*, 2011; Larque *et al.*, 2003; Ghaeni and Ghahfarokhi, 2013; Chen and Liu, 2020). Therefore, a low TI value indicates that consuming foods or products is beneficial for cardiovascular health (CVH) (Chen and Liu, 2020). Low AI and TI values are preferred in lipids (Luczynska and Beata Paszczyk, 2019). The lower the AI and TI values, the more beneficial the product is for health, which is related to the positive effect of MUFA and PUFA on the cardiovascular system (Turan *et al.*, 2007; Murzina *et al.*, 2022). The FLQ index shows the quality of the food source of lipids. The calculation of FLQ, taking into account the content of EPA and DHA that are rapidly oxidized, it is recommended to use this indicator to determine the "freshness" of the product (Abrami *et al.*, 1992; Senso *et al.*, 2007; Murzina *et al.*, 2022). The FLQ values of all samples were calculated to be in the range of 4-8.

The hypocholesterolemic/hypercholesterolemic (H/H) ratio is an index used in the fatty acid profile of lamb meat first proposed by Santos-Silva *et al.* (2002) (Chen and Liu, 2020). Santos-Silva *et al.* (2002) have developed H/H as a new index to evaluate the effect of fatty acid composition on cholesterol in lambs due to low polyunsaturated fatty acid (PUFA) to saturated fatty acid (SFA) ratio based on high saturated (SAT) fatty acid content. H/H, which is a fundamental research on the regulation of FA in the diet and plasma LDL-C, characterizes the relationship between hypocholesterolemic fatty acids (cis-C18:1 and PUFA) and hypercholesterolemic FA (Dietschy, 1998; Chen and Liu, 2020). Santos-Silva *et al.* (2002) have stated that since C12:0 was not detected in lambs, the formula only included C14:0 and C16:0 in hypercholesterolemic fatty acids (Chen and Liu, 2020). The H/H index was calculated between 3-4,23. UI is used to indicate the degree of unsaturation in lipids, and is calculated as the product of the sum of the percentage of each unsaturated FA and the number of double bonds in that FA (Logue, 2000; Chen and Liu, 2020). Unlike UFA and PUFA, different unsaturated fatty acids have different weights within the UI. It is generally stated that UI more comprehensively reflects the ratio of different unsaturation degrees of fatty acids in the total fatty acid composition of a food component (Chen and Liu, 2020). For example, the UI values of some seaweed species range from 45 to 368.68, with *Gracilaria changii*

having a UI of 368.68 (Chan and Matanjun, 2017). In our study, the UI values were determined to be between 113-121, and it was seen that the unsaturation index (UI) of the species was good. One of the main purposes of giving such index calculations is to obtain more comparable data in terms of the relationship between fatty acids, health and quality. In this way, more comparable results can be obtained between studies. The polyene index (PI) has been used as a measure of PUFA damage (Lubis and Buckle, 1990; Küçükgülmez *et al.*, 2018). Šimat *et al.* (2015) have stated that the decrease in PI values of bogue (Boops boops Linnaeus, 1758) fish during cold storage is a sign of the breakdown of PUFAs, and that with the decrease in PI, primary and secondary oxidation products, i.e. PV (peroxide value) and TBA components, are concentrated.

Table 2. LQI results in waste skin and bones of cultured fish using the percentage results (%) of fatty acids

	OM Skin	OM Bone	SP Skin	SP Bone	DL Skin	DL Bone
AI	0,23	0,18	0,22	0,22	0,18	0,24
TI	0,71	0,98	0,71	0,81	0,90	0,80
FLQ	8,39	4,19	9,17	8,15	4,89	7,57
h/H	2,93	4,23	3,00	2,95	4,15	2,85
HPI	3,58	4,75	3,70	3,73	4,69	3,49
UI	121	113	125	118	118	116
PI	0,54	0,37	0,63	0,59	0,42	0,49
DFA	81	87	82	83	87	81
OFA	16	11	15	14	12	15

OM: and *Oncorhynchus mykiss* (Rainbow trout), SP: *Sparus aurata* (Sea bream), DL: *Dicentrarchus labrax* (Sea Bass)

ALA is known as an essential fatty acid because it cannot be synthesized from saturated and polyunsaturated fatty acids (PUFAs) in humans. The importance of n-3 fatty acids has been increasingly seen in recent years. One of the important focus points of nutrition has been to determine the individual amount of n-3 fatty acid needed to ensure adequate nutrition and prevent deficiency. It is stated that significant progress has been made in determining recommendations for n-3 fatty acids in the prevention and treatment of chronic diseases in recent times (Gebauer *et al.*, 2006). The National Heart, Lung, and Blood Institute Family Heart Study (Djoussé *et al.*, 2001), the Nurses' Health Study (Hu *et al.*, 1999), and the Health Professionals Follow-up Study (Ascherio *et al.*, 1996) have shown that there is a positive relationship between ALA and cardiovascular disease (CVD) risk (Gebauer *et al.*, 2006). Many significant studies in cardiovascular disease have been shown that consumption of EPA, DHA, and fish has a cardioprotective effect (Hu *et al.*, 2002; Albert *et al.*, 1998; Lemaitre *et al.* 2003; Daviglus *et al.* 1997; Orenca *et al.*, 1996; Zhang *et al.*, 1999; Gebauer *et al.*, 2006). It has been stated that fish oil can be used as a potential supplement to improve the severity of some skin disorders such as skin cancer, allergies, dermatitis, cutaneous wounds and melanogenesis (Huang *et al.*, 2018).

According to The Australian National Health and Medical Research Council (NHMRC, 2006), recommendations for daily intake of some fatty acids and groups based on life stage and gender are given in Table 3. In our study,  $\omega$ -3 and  $\omega$ -6 polyunsaturated fatty acids were found to be between 1.57-5.64 and 9.07-18.97 g/100g, respectively. Linoleic acid (LA) and alpha-linolenic acid (ALA) were determined to be between 4.64-9.33 and 0.11-0.30 g/100g, respectively. Based on our results, it is seen that the amounts of  $\omega$ -3,  $\omega$ -6, and Linoleic acid (LA) fatty acids are over the daily intake

amounts despite the low levels of  $\alpha$ -linolenic acid (ALA) in fish skin and bones based on 100g of dry weight (DWC).

Table 3. Recommendations of NHMRC (2006) for daily consumption of some fatty acids and groups according to life stage and gender

	<b>Infants</b>					
	0–6 months			7–12 months		
$\omega$ -6 polyunsaturated fats	4.4 g/day			0.5 g/day		
$\omega$ -3 polyunsaturated fats	4.6 g/day			0.5 g/day		
<b>Children, adolescents &amp; adults</b>						
	<b>Boys</b>		<b>Girls</b>		<b>Adults 19+ yr</b>	
	9–13 yr	14–18 yr	9–13 yr	14–18 yr	Men	Women
Linoleic acid (LA)	10 g/day	12 g/day	8 g/day	8 g/day	13 g/day	8 g/day
$\alpha$ -linolenic acid (ALA)	1.0 g/day	1.2 g/day	0.8 g/day	0.8 g/day	1.3 g/day	0.8 g/day
Total LC $\omega$ -3 (DHA+EPA+DPA)	70 mg/day	125 mg/day	70 mg/day	85 mg/day	160 mg/day	90 mg/day

Bruni *et al.* (2021) investigated n-3 fatty acids in rainbow trout skin. In the study, it was found  $\omega$ -3 fatty acids between 10 and 22 % (as percentage of total FAMES). In addition, EPA and DHA were obtained between 3.12 and 6.85, and 5.97 and 13.42%, respectively. In another study, Sae-leaw and Benjakul (2014) investigated the fatty acid profile, lipid hydrolysis and oxidation, fish odor, and volatile compounds of sea bass (*Lates calcarifer*) skin during 18 days of frozen storage. In the study, SAT, MUFA, PUFA, EPA and DHA were determined as 35.98, 29.57, 27.90, 3.04 and 9.82 g/100 g lipids in sea bass skin, respectively. In our study, SAT, MUFA, PUFA, EPA and DHA were determined as 13.38, 4.49, 11.54, 0.34 and 1.10 g/100g dry weight composition (DWC) in sea bass skin, respectively. When the DWC results in our study were calculated as g/100 g lipid according to Sae-leaw and Benjakul (2014), our results were calculated as 43.15, 14.47, 37.21, 1.09 and 3.55 g/100 g lipid, respectively. As a result, it can be said that the results we obtained are in similar agreement with the literature data.

## Conclusions

The determination of fatty acid components is an important criterion for evaluating the quality of fat components. Fatty acids are important for human health and most fatty acids can be synthesized in the body, but the human body does not have the necessary enzymes to produce the two basic fatty acids EPA and DHA. Therefore, these fatty acids must be taken in food. Today, the quality and quantity of food has become one of the most important issues. The quality of nutrition is important for human health. Alternative important fatty acids in food sources, such as seafood, are known to have a rich nutritional composition, including essential amino acids, unsaturated fatty acids, vitamins, minerals, and antioxidants, as well as the necessary nutrients for human life. The demand for seafood is met through aquaculture and fishing. In this study, the fatty acids were determined in skin and bone of cultured *Sparus aurata* (Sea bream), *Dicentrarchus labrax* (Sea Bass) and

*Oncorhynchus mykiss* (Rainbow trout) obtained Canakkale fish market using Gas Chromatography (GC). In addition, lipid quality index parameters were investigated in skin and bones of fish. The main purpose of this study was to investigate whether fish skin and bones, known as waste, can be used as a qualified lipid resource. The results obtained were calculated both in percentage and in gram amounts per 100g portion. In this study, EPA and DHA were in the range from 1 to 3.75% and from 3 to 6%, respectively. The amount of unsaturated fatty acids was calculated as 15 to 33 g/100 g as DWC. As a result of the determination of fatty acids, it was achieved that cultured skin and bones of fish can be considered as a high-quality lipid source. When evaluated with these measures, cultured fish skin and bones is seen to be rich in EPA, DHA and omega-3 fatty acids. In addition, the lipid quality index showed that the fat quality is high. In particular, the Atherogenicity Index (AI) and the Thrombogenicity Index (TI) are able to show that the fat type has good quality characteristics.

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