

Determination of lipid quality and mercury levels of sardine and rainbow trout cooked with different methods

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Abstract: This study aimed to investigate the effects of baking and pan-frying methods on the lipid quality and mercury (Hg) levels of two important fish species in Türkiye, namely, fileted sardine (*Sardina pilchardus*) and rainbow trout (*Oncorhynchus mykiss*). The results revealed that sardines significantly decreased n-3 fatty acids depending on the cooking process, while the best n-6/n-3 ratio was observed in baked sardines, with higher rates found in pan-fried fish. Notably, pan-fried rainbow trout cooked with butter showed the highest atherogenic index (AI) of 0.71 ± 0.32 and thrombogenic index (TI) of 0.61 ± 1.43 , as well as a hypocholesterolemic/hypercholesterolemic index (HH) of 0.79 ± 0.17 . Conversely, fried sardines exhibited lower atherogenic and thrombogenic scores, with fried sardines cooked in sunflower oil having a hypocholesterolemic/hypercholesterolemic index of 4.85 ± 0.3 . There were no significant variations in Hg content between raw and cooked fish. However, when compared to the raw control, the rise in Hg content for baked fish was substantial ($p < 0.05$) (baked rainbow trout 0.18 mg/kg and sardine 0.29 mg/kg). The decrease in FAs (Fatty Acids) due to cooking methods can be ordered as follows: Raw sardine > baked > fried. Conversely, the increase in FAs due to the cooking methods can be ordered as follows: fried > baked > raw sardine. Baked rainbow trout had much lower levels of palmitic acid, stearic acid, and myristic acid than the raw material. However, these levels were significantly increased in rainbow trout cooked in a frying pan. Additionally, oleic acid levels in fried rainbow trout were detected to be lower than in other preparations, whereas they were equivalent in baked rainbow trout.

Keywords: Fatty acids composition, pan-frying; baking, lipid quality, mercury, sardine, rainbow trout

INTRODUCTION

Because of its high polyunsaturated fatty acids (PUFAs) content with a significant amount of omega 3, which is not naturally present in the human body, and its low content of saturated fatty acids (SFA), fish is preferred for consumption. (Erdem and Dinçer, 2019). Additionally, long-chain n-3 PUFA is abundant in the lipids of fish meat, and seafood (Erdem et al., 2020). They are key cell membrane constituents that contribute to a variety of membrane activities. (EFSA, 2012a). It is well known that the amount of nutrients and toxins accumulated in the body depends on the species and the amount consumed. Heavy metals, such as lead, cadmium, and mercury, can accumulate in the tissues of fish, especially in their flesh. These metals can come from a variety of sources, including pollution, industrial waste, and contaminated waterways. Mercury is one of the most concerning heavy metals found in fish, as it can have toxic effects on the nervous system, particularly in young children and pregnant women. Certain types of fish, such as large predatory fish like swordfish and sharks, tend to have higher levels of mercury due to their position in the food chain. The abundance of heavy metals and mercury in fish flesh is a significant public health concern, and guidelines exist to help consumers make informed decisions about which types of fish are safe to eat and how often to

consume them. Cooking fish and seafood before eating is a common practice. This heat treatment is a necessary precaution to ensure that the food offers the desired texture, flavor, and color during the cooking process or food preparation, in addition to ensuring food cleanliness and safety (Erdem and Dinçer, 2019).

The effects of several cooking methods on nutritional quality, particularly fatty acid (FA) profiles and mercury concentration have recently been examined (Karimian-Khosroshahi et al., 2016; Zhang et al., 2019; Islam et al., 2020). Methods of cooking, including frying, boiling, baking, microwaving, and steaming, affect the composition of various fatty acids, with PUFAs being altered by these cooking methods and fish species (Farag, 2013; Flaskerud et al., 2017; Alexi et al., 2019). In Türkiye, sunflower oil, which contains approximately 71% polyunsaturated fatty acids (PUFA), is popular culinary oil (Demirtaş Erol et al., 2022). Sardines (*Sardina pilchardus*) and rainbow trout (*Oncorhynchus mykiss*) are popular types of fish consumed in many parts of the world, including Türkiye. Sardines are a staple food in Mediterranean countries, and they are also commonly consumed in Asia, South America, and Africa. On the other hand, rainbow trout is widely consumed in Europe and North America.

In Türkiye, sardines are a popular seafood item, especially in the coastal regions. They are typically grilled or fried and served with salad or bread. Rainbow trout are also consumed in Türkiye, particularly in the Black Sea region, where it is abundant in the local rivers, and it is mostly served pan-fried with butter. These two species were selected to investigate possible changes induced by different cooking methods.

MATERIALS AND METHODS

Fish

Sardines (*Sardina pilchardus*) with an approximate weight of $94.6 \text{ g} \pm 10.2$ and length of $15.6 \text{ cm} \pm 1.4$ were freshly bought from the fish market in Buca, İzmir, Türkiye. Farm-raised rainbow trout (*Oncorhynchus mykiss*) with an approximate weight of $250 \text{ g} \pm 35$ and length of $20.1 \text{ cm} \pm 1.4$ were obtained from an aquaculture facility in Manisa, Türkiye. The fish were transferred to the laboratory under a cold chain. Afterward, the fish, with their skins on, were filleted and cooked depending on cooking methods.

Fish heat treatment

Cooking techniques were based on previously published methods (Flaskerud et al., 2017; Farag, 2013). For this study, pan-frying and baking were selected as the cooking methods. In Türkiye, rainbow trout is a widely consumed freshwater fish species, and it is often prepared by frying. However, in Turkish households, the use of saturated fats for pan-frying is being increasingly replaced by healthier unsaturated vegetable oils, especially olive oil. Some families still prefer using butter for pan-frying (Bilgin et al., 2010). Therefore, sardine is a widely caught and consumed marine fish species. The temperature and time used in this study were chosen based on references used for cooking methods. However, there have been numerous studies on the effect of different cooking methods on sardines and rainbow trout. The goal of this study, different from other research, is to reveal the effect of different cooking techniques on both mercury and fatty acids.

Sardine fillets were pan-fried in sunflower oil for 10 minutes on both sides at 180°C . The rainbow trout fillets were pan-fried in butter for 12 minutes on each side at 180°C and then gently drained for approximately 2 minutes. For the baking process, sardines were placed in the oven (Öztiryakiler, OKFE 101, İzmir, Türkiye) and baked at 180°C for 22 minutes, while rainbow trout fillets were baked for 30 minutes at the same temperature. The cooking procedure was considered complete when a quartz electronic thermometer indicated that the fillet's interior temperature ranged between 60 and 70°C . Once the necessary temperature was obtained for all cooking methods, the samples were cooled and tested.

Fatty acid analysis

Fatty acid analyses were carried out using the IUPAC II.D.19 method (IUPAC, 1979). Fatty acids of the anchovy and anchovy oil were analyzed using a Perkin Elmer Auto system XL Gas Chromatograph equipped with SP-2330 and a flame

ionization detector (FID). Separation of fatty acid methyl esters was achieved on a fused silica capillary column ($30 \text{ m} \times 0.25 \text{ mm} \times 0.20 \mu\text{m}$ film thicknesses) the oven temperature was 120°C for 2 min, and programmed to 220°C at a heating rate of $5^\circ\text{C}/\text{min}$, then held for 15 min. The injector and detector temperatures were maintained at 240°C and 250°C , respectively. The carrier gas was helium 10psi with a split ratio of 1/50. The air and hydrogen pressure were 338 ml/min and 45 ml/min respectively. Fatty acids were identified by comparing the retention times of fatty acid methyl esters (FAME) with a standard 37-component FAME mixture (Supelco- Catalog No:18919-1Amp.) Results were expressed as the percentage of each fatty acid concerning the total fatty acids. The GC analyses were performed in triplicate, and the results were expressed as % of total FAME area as the mean value of a percentage.

Lipid quality indices

The thrombogenic index (TI) and atherogenic index (AI) were calculated due to FA composition by using the method of Ulbricht and Southgate (1991). The hypocholesterolemic/hypercholesterolaemic ratio (HH) was calculated according to fatty acid composition by using the method Santos-Silva et al. (2002) using the following equations:

$$AI = \frac{12:0 + (4 \times 14:0) + 16:0}{\sum UFA}$$

$$TI = \frac{14:0 + 16:0 + 18:0}{(0.5MUFA) + (0.5n-6PUFA) + (3n-3PUFA) + ((n-3PUFA)/(n-6PUFA))}$$

$$HH = \frac{18:1n-9 + 18:2n-6 + 20:4n-6 + 18:3n-3 + 20:5n-3 + 22:5n-3 + 22:6n-3}{12:0 + 14:0 + 16:0}$$

¹MUFA: monounsaturated fatty acid

Determination of total mercury (Hg) content

For the quantitative analyses of total mercury (Hg), fish samples were digested. Wet samples and HNO_3 were taken in the tube and digested according to the program of eicosapentaenoic acid (EPA) Methods (1994). After digestion, each sample was transferred to a 50 ml volumetric flask and filled up to the mark with deionized water. The sample was filtered and further diluted by four times to be analyzed by ICP-MS (Agilent 7500CE, USA). The standard solutions were prepared by diluting the required amount of the solution from the stock solution, manufactured by Agilent, Germany.

Statistical analysis

Statistical analysis was performed using IBM SPSS (statistical package for the social sciences) Statistics 22.0 and expressed as mean \pm SD of the three replicated cooking processes. To define the significance of differences in proximate value, fatty acid content, and nutritional quality before and after cooking, analysis of variance ANOVA) using one way followed by Tukey's significant difference test ($p < 0.05$). All data are expressed as mean \pm standard deviation. Principal component analysis (PCA) explored differences in the three groups' compositions.

RESULTS

FA composition of pan-fried and baked sardine

Fatty acids were classified as SFA, MUFA, and PUFA, and a total of 33 fatty acids were examined. Different cooking techniques resulted in various alterations in the fatty acid composition. The fatty acids of raw, fried, and baked sardines are shown in Table 1. All cooking methods reduced the total SFA, HUFA, and n-3 PUFA, while they increased total MUFA, PUFA, and n-6 PUFA in baked and fried sardines when compared to raw sardines. Palmitic acid (C16:0) was the major

constituent of SFA. Myristic, palmitic, palmitoleic, stearic, linoleic, and eicosapentaenoic acids decreased in baked and fried sardines compared to raw sardines. The decrease in FAs due to cooking methods can be ordered as follows: Raw sardine > baked > fried. Among the MUFAs, oleic acid was the most abundant in sardines. Additionally, oleic, linoleic, and docosahexaenoic acids increased in baked and fried sardines compared to raw sardines. The increase in FAs due to the cooking methods can be ordered as follows: fried > baked > raw sardine. PUFA in cooked fish significantly differed from those in raw fish ($p < 0.001$).

Table 1. Fatty acids (%) profile of cooked by different methods and raw sardine

Fatty acids composition	Raw	Baked	Fried
Caproic acid (C6:0)	0.02 ± 0.001	nd	nd
Caprylic acid (C8:0)	0.01 ± 0.005	nd	nd
Capric acid (C10:0)	0.03 ± 0.02	nd	nd
Undecanoic acid (C11:0)	0.01 ± 0.002	nd	nd
Lauric acid (C12:0)	0.13 ± 0.02 ^a	0.08 ± 0.05 ^{ab}	0.04 ± 0.02 ^b
Tridecanoic acid (C13:0)	0.05 ± 0.01 ^a	0.03 ± 0.01 ^a	0.02 ± 0.02 ^a
Myristic acid (C14:0)	5.12 ± 0.1 ^a	3.36 ± 0.04 ^b	1.99 ± 0.22 ^c
Myristoleic acid (C14:1)	0.02 ± 0.01	nd	nd
Pentadecanoic acid (C15:0)	0.86 ± 0.01 ^a	0.57 ± 0.03 ^b	0.31 ± 0.02 ^c
Palmitic acid (C16:0)	22.95 ± 0.05 ^a	17.02 ± 0.08 ^b	12.38 ± 0.12 ^c
Palmitoleic acid (C16:1)	5.58 ± 0.42 ^a	3.77 ± 0.73 ^b	2.28 ± 0.67 ^c
Heptadecanoic acid (C17:0)	0.62 ± 0.04 ^a	0.44 ± 0.06 ^b	0.23 ± 0.09 ^c
Stearic acid (C18:0)	4.86 ± 0.04 ^a	4.14 ± 0.06 ^b	3.57 ± 0.44 ^c
Elaidic acid (C18:1n9t)	0.15 ± 0.02 ^a	0.06 ± 0.01 ^b	0.04 ± 0.01 ^b
Oleic acid (C18:1n9c)	14.87 ± 0.13 ^a	20.5 ± 0.5 ^b	26.01 ± 2.99 ^c
Linoleic acid (C18:2n6c)	2.39 ± 0.01 ^a	19.71 ± 0.28 ^b	33.67 ± 1.33 ^c
Arachidic acid (C20:0)	0.77 ± 0.02 ^a	0.59 ± 0.03 ^b	0.43 ± 0.13 ^c
γ-Linolenic acid (C18:3n3)	0.13 ± 0.03 ^a	0.09 ± 0.04 ^{ab}	0.05 ± 0.01 ^b
11-Eicosenoic acid (C20:1)	1.63 ± 0.03 ^a	1.04 ± 0.04 ^b	0.7 ± 0.17 ^a
α-Linolenic acid (C18:3n3)	1.9 ± 0.1 ^a	1.31 ± 0.07 ^b	0.8 ± 0.4 ^c
Heneicosanoic acid (C21:0)	0.03 ± 0.01 ^a	0.02 ± 0.02 ^a	0.01 ± 0.05 ^a
Eicosadienoic acid (C20:2)	3.75 ± 0.05 ^a	2.58 ± 0.12 ^b	1.6 ± 0.7 ^c
Behenic acid (C22:0)	0.25 ± 0.02 ^a	0.41 ± 0.09 ^b	0.5 ± 0.05 ^b
8,11,14-Eicosatrienoic acid (C20:3n6)	0.09 ± 0.01 ^a	0.06 ± 0.04 ^a	0.04 ± 0.01 ^a
Erucic acid (C22:1n9)	0.26 ± 0.01 ^a	0.17 ± 0.06 ^b	0.11 ± 0.01 ^b
11,14,17-Eicosatrienoic acid (C20:3n3)	0.17 ± 0.02 ^a	0.12 ± 0.04 ^{ab}	0.07 ± 0.01 ^b
Arachidonic acid (C20:4n6)	0.4 ± 0.08 ^a	0.34 ± 0.04 ^a	0.18 ± 0.03 ^b
13,16-Docosadienoic acid (C22:2)	0.86 ± 0.04 ^a	0.62 ± 0.04 ^b	0.38 ± 0.04 ^c
Lignoceric acid (C:24:0)	0.09 ± 0.01 ^a	0.15 ± 0.05 ^{ab}	0.18 ± 0.03 ^b
5,8,11,14,17-Eicosapentaenoic acid (C20:5n3)	8.06 ± 0.04 ^a	5.9 ± 0.4 ^b	3.72 ± 0.24 ^c
Nervonic acid (C24:1)	0.53 ± 0.03 ^a	0.4 ± 0.07 ^b	0.23 ± 0.09 ^c
7,10,13,16,19-Docosapentaenoic acid (C22:5n3)	0.9 ± 0.2 ^a	0.64 ± 0.11 ^a	0.4 ± 0.05 ^b
4,7,10,13,16,19-Docosahexaenoic acid (c22:6n3)	11.19 ± 0.31 ^a	8.14 ± 0.36 ^b	5.13 ± 0.18 ^c
Σ SFA	35.8 ± 0.1 ^a	26.81 ± 0.02 ^b	18.53 ± 0.02 ^c
Σ MUFA	23.04 ± 0.04 ^a	25.94 ± 0.06 ^b	35.83 ± 0 ^c
Σ HUFA	19.25 ± 0.05 ^a	14.04 ± 0.02 ^b	7.39 ± 0.02 ^c
Σ PUFA	29.84 ± 0.16 ^a	39.51 ± 0.37 ^b	39.85 ± 0.01 ^b
Σ PUFA (n-3)	22.35 ± 0.01 ^a	16.2 ± 0.15 ^b	10.6 ± 0.06 ^c
Σ PUFA (n-6)	2.88 ± 0.02 ^a	20.11 ± 0.11 ^b	27.87 ± 0.01 ^c

Means in the same line with the same letter do not differ significantly at the level of 0.05 significance. nd: not detected; SFA: saturated fatty acid; MUFA: monounsaturated fatty acid; PUFA, polyunsaturated fatty acid; HUFA, highly unsaturated fatty acid. n:3 (arithmetic mean±SD)

Fatty acids composition of pan-fried and baked rainbow trout

The fatty acid composition of rainbow trout was given in Table 2. Palmitic acid, stearic acid, and myristic acid were dramatically reduced in baked rainbow trout compared to raw material. However, they increased significantly in pan-fried rainbow trout. Additionally, the oleic acid content in baked rainbow trout remained similar to the raw one, while it was lower in fried rainbow trout compared to the other cooking methods.

Total SFA was the highest in fried rainbow trout. Total MUFA was the highest in raw rainbow trout but close to that of baked rainbow trout. Total HUFA, PUFA, n-3 PUFA, and n-6 PUFA were the highest in baked rainbow trout. Total n-3 PUFA was higher in baked rainbow trout than in raw and fried ones. The order for the n-3 PUFA content of rainbow trout is as follows: baked > raw rainbow trout > fried. Linoleic, eicosapentaenoic, and docosahexaenoic acids were higher in baked rainbow trout than in the other cooking methods.

Table 2. Fatty acids (%) profile of cooked by different methods and raw rainbow trout

Fatty acids composition	Raw	Baked	Fried
Caproic acid (C6:0)	0.13 ± 0.13 ^a	nd	0.72 ± 0.07 ^b
Caprylic acid (C8:0)	0.08 ± 0.08 ^a	nd	0.65 ± 0.07 ^b
Capric acid (C10:0)	nd	nd	0.68 ± 0.04
Undecanoic acid (C11:0)	0.04 ± 0.04 ^a	nd	0.13 ± 0.1 ^b
Lauric acid (C12:0)	0.07 ± 0.07 ^a	0.04 ± 0.02 ^a	0.76 ± 0.24 ^b
Tridecanoic acid (C13:0)	0.02 ± 0.02 ^a	0.01 ± 0.01 ^a	nd
Myristic acid (C14:0)	4.42 ± 0.81 ^a	1.94 ± 0.17 ^b	5.98 ± 0.53 ^c
Myristoleic acid (C14:1)	nd	0.02 ± 0.02 ^a	0.08 ± 0.02 ^b
Pentadecanoic acid (C15:0)	0.32 ± 0.32 ^a	0.15 ± 0.08 ^a	0.51 ± 0.06 ^a
Palmitic acid (C16:0)	24.45 ± 1.87 ^a	11.43 ± 0.89 ^b	29.15 ± 0.3 ^c
Palmitoleic acid (C16:1)	3.69 ± 0.27 ^a	2.91 ± 0.14 ^b	1.66 ± 0.16 ^c
Heptadecanoic acid (C17:0)	0.33 ± 0.33 ^a	0.14 ± 0.05 ^a	0.41 ± 0.02 ^a
Stearic acid (C18:0)	7.14 ± 1.27 ^a	3.38 ± 0.46 ^b	9.21 ± 0.23 ^c
Elaidic acid (C18:1n9t)	0.1 ± 0.1 ^a	0.12 ± 0.02 ^a	0.04 ± 0.01 ^b
Oleic acid (C18:1n9c)	32.61 ± 2.6 ^a	33.12 ± 2.09 ^a	21.26 ± 2.79 ^b
Linoleic acid (C18:2n6c)	6.57 ± 0.99 ^a	24.47 ± 0.5 ^b	5.05 ± 0.9 ^a
Arachidic acid (C20:0)	0.37 ± 0.37 ^a	0.21 ± 0.11 ^a	0.44 ± 0.11 ^a
γ-Linolenic acid (C18:3n3)	0.08 ± 0.08 ^a	0.29 ± 0.05 ^b	0.02 ± 0.02 ^a
11-Eicosenoic acid (C20:1)	2.28 ± 1.28 ^a	2.15 ± 0.05 ^a	0.89 ± 0.09 ^a
α-Linolenic acid (C18:3n3)	0.89 ± 0.11 ^a	3.32 ± 0.18 ^b	0.44 ± 0.22 ^b
Heneicosanoic acid (C21:0)	0.02 ± 0.01 ^a	0.01 ± 0.006 ^a	nd
Eicosadienoic acid (C20:2)	0.61 ± 0.24 ^a	1.56 ± 0.09 ^b	0.25 ± 0.27 ^a
Behenic acid (C22:0)	0.18 ± 0.05 ^a	0.2 ± 0.15 ^a	0.52 ± 0.13 ^b
8,11,14-Eicosatrienoic acid (C20:3n6)	0.16 ± 0.07 ^a	0.45 ± 0.2 ^b	nd
Erucic acid (C22:1n9)	0.28 ± 0.28 ^{ab}	0.28 ± 0.08 ^a	0.11 ± 0.05 ^b
11,14,17-Eicosatrienoic acid (C20:3n3)	0.09 ± 0.09 ^a	0.32 ± 0.14 ^a	nd
Arachidonic acid (C20:4n6)	0.09 ± 0.06 ^a	0.4 ± 0.04 ^b	0.04 ± 0.005 ^a
13,16-Docosadienoic acid (C22:2)	0.16 ± 0.16 ^a	0.6 ± 0.2 ^b	0.03 ± 0.01 ^c
Lignoceric acid (C:24:0)	0.07 ± 0.1 ^a	0.08 ± 0.02 ^a	nd
5,8,11,14,17-Eicosapentaenoic acid (C20:5n3)	0.45 ± 0.09 ^a	1.75 ± 0.12 ^b	0.25 ± 0.11 ^a
Nervonic acid (C24:1)	0.27 ± 0.25 ^a	0.21 ± 0.12 ^a	0.09 ± 0.01 ^a
7,10,13,16,19-Docosapentaenoic acid (C22:5n3)	0.32 ± 0.14 ^a	0.63 ± 0.3 ^a	0.37 ± 0.12 ^a
4,7,10,13,16,19-Docosahexaenoic acid (c22:6n3)	1.25 ± 0.14 ^a	3.97 ± 0.05 ^b	0.83 ± 0.11 ^c
Σ SFA	37.64 ± 2.66 ^a	17.59 ± 1.36 ^b	49.16 ± 3.47 ^c
Σ MUFA	39.23 ± 2.01 ^a	38.81 ± 1.42 ^a	24.13 ± 2.23 ^b
Σ HUFA	1.7 ± 0.07 ^a	5.72 ± 1.53 ^b	1.08 ± 1.08 ^a
Σ PUFA	10.67 ± 0.91 ^a	37.76 ± 1.49 ^b	7.28 ± 0.57 ^c
Σ PUFA (n-3)	3.08 ± 0.67 ^a	10.28 ± 2.52 ^b	1.91 ± 0.3 ^c
Σ PUFA (n-6)	6.82 ± 1.54 ^a	25.32 ± 1.8 ^b	5.09 ± 0.19 ^a

Means in the same line with the same letter do not differ significantly at the level of 0.05 significance. nd: not detected; SFA: saturated fatty acid; MUFA: monounsaturated fatty acid; PUFA, polyunsaturated fatty acid; HUFA, highly unsaturated fatty acid. n:3 (arithmetic mean±SD)

In fried sardines cooked with sunflower oil, a significant decrease ($p < 0.05$) was observed in myristic, palmitic,

palmitoleic, stearic, linolenic, arachidic, and docosahexaenoic acid (DHA). However, there was a significant increase

($p < 0.05$) in oleic and linoleic acid. The DHA content was decreased in all fried groups.

Indices of lipid quality

These indices take into account the many consequences that fatty acids may have on human health, especially the likelihood that atherosclerosis and/or thrombus formation may become more common. The atherogenic index (AI) and thrombogenicity index (TI) were respectively found 0.71 ± 0.32 and

0.61 ± 1.43 to be highest in butter-fried rainbow trout.

Therefore, the hypocholesterolemic / hypercholesterolemic (HH) index of rainbow trout fried in butter (0.79 ± 0.17) was found to be the best in terms of nutritional quality. The atherogenic and thrombogenic index were determined lower in all groups of sardines. The hypocholesterolemic / hypercholesterolemic index of fried sardine (4.85 ± 0.3) in sunflower oil was found to be the best in terms of nutritional quality (Table 3).

Table 3. Lipid quality indices in sardine and rainbow trout after cooked by different methods

	Rainbow trout AI	Rainbow trout TI	Rainbow trout HH	Sardine AI	Sardine TI	Sardine HH
Raw	0.29 ± 0.19^a	0.23 ± 0.02^a	1.46 ± 0.54^a	0.06 ± 0.01^a	0.02 ± 0.01^a	1.4 ± 0.23^a
Baked	0.01 ± 0.16^a	0.01 ± 0.02^b	5.05 ± 0.71^b	0.02 ± 0.02^b	0.01 ± 0.01^a	2.76 ± 0.14^b
Fried	0.71 ± 0.32^b	0.61 ± 0.43^c	0.79 ± 0.17^a	0.01 ± 0.02^b	0.01 ± 0.00^a	4.85 ± 0.3^c

Means in the same column with the same letter do not differ significantly at the level of 0.05 significance. n:3 (arithmetic mean \pm SD)

Mercury (Hg) content

The content of Hg is given in Table 4. The Hg content of raw rainbow trout was found 0.08 mg/kg. This value is 0.11 mg/kg in raw sardine. In both fish baking increased the Hg levels than frying. Fried rainbow trout was significantly different

from raw and baked ones ($p < 0.05$).

Therefore, baked sardine was significantly different from raw and fried one. Fish had higher Hg contents after cooking, according to several studies (Girard et al., 2018; Burger et al., 2003).

Table 4. Mercury (Hg) content (mg/kg) of raw and cooked rainbow trout and sardine

Rainbow trout			Sardine		
Raw	Baked	Fried	Raw	Baked	Fried
0.08 ± 0.03^a	0.18 ± 0.07^a	0.08 ± 0.02^b	0.11 ± 0.1^a	0.29 ± 0.04^b	0.15 ± 0.03^a

Means in the same line in the same group with the same letter do not differ significantly at the level of 0.05 significance. n: 3 (arithmetic mean \pm SD)

DISCUSSION

Due to the association between these fatty acids and health benefits, the quantity of n-3 PUFAs in fish, particularly EPA and DHA, can be used to determine nutritional quality.

Our findings are consistent with those of Karimian-Khosroshahi et al. (2016). The nutritional value of rainbow trout was estimated by studying the effects of baking and pan-frying. The study examined the chemical composition, lipid quality indexes, fatty acid profile, and mercury levels of rainbow trout. Ideal n-6/n-3 human nutrition values are considered to be 1-1.5 or less (Larrieu and Layé, 2018). Baked sardines represent the optimum n-6/n-3 ratio, whereas these rates are quite high in pan-fried fish.

Eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) have been shown to have numerous health benefits, including reducing inflammation, lowering blood triglyceride levels, and reducing the risk of heart disease. According to the AHA, adults should aim to consume at least two servings of fatty fish per week, which can provide about 500 milligrams of EPA and DHA for daily intake. The European Food Safety Authority (EFSA) suggests a range of 250–500 mg/day based on cardiovascular risk concerns for European adults (Kris-Etherton et al., 2002).

The increased level of SFA in fried rainbow trout fillets is assumed to be caused by the butter used. Dairy products, in

particular butter, have been considered to increase the risk for cardiovascular diseases in humans because, in comparison to other lipid sources, they contain a higher proportion of lauric, myristic, and palmitic acids and a lower proportion of unsaturated fatty acids (Sacks and Katan, 2002) proposed an atherogenic index (AI) for lipids as a dietary risk indicator for cardiovascular disease. Sunflower oil contains approximately 15% saturated, and 85% unsaturated fatty acid and consists of 14–43% oleic and 44–75% linoleic acids in its unsaturated fatty acid content (Akkaya, 2018).

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Effects of the cooking methods on lipid quality were related to the containing of meat dehydration, fat migration to the frying oil, and oil penetration to meat. Frying had the greatest impact on lipid quality, but its impact varied depending on the species.

Due to their ability to prevent the development of plaque and lower levels of cholesterol, phospholipids, and esterified fatty acids, unsaturated fatty acids are thought to be anti-atherogenic. As a result, consuming meals or goods with a lower AI can lower LDL-C and total cholesterol in blood plasma values. HH values for shellfish range from 1.73 to 4.75, except for *Loxechinus albus*. For fish, the values varied from 1.54 to 4.83, except for *Opisthonema oglinum*, which has an HH value of 0.87. For dairy products and meat, the ranges are 1.27–2.786, and 0.32–1.29, respectively (Chen and Liu, 2020). Fish consumption is the primary pathway through which people are exposed to mercury. Seafood is widely used in traditional cuisines around the world even though it quickly bioaccumulates mercury (Hg). Only a small number of previous studies on Hg in cooked seafood took into account both MeHg and Hg(II); the majority concentrated on total mercury (Liao et al., 2019). Although Hg levels (Burger et al., 2003; Khansari et al., 2005; Kalogeropoulos et al., 2012; Jadán-Piedra et al., 2017; Liao et al., 2019; Dahl et al., 2020) are typically highest in well-distributed fish organs like the liver, spleen, and kidney (Sandheinrich and Wiener, 2011; Matos et al., 2015) the greatest pool of Hg in fish is found in the muscle. In the fish muscle, > 95% of the Hg(II) is present as MeHg (Bloom, 1992).

It has been hypothesized that this has something to do with weight loss brought on by moisture and fat loss during cooking (Morgan et al., 1997). Multiple studies on mercury in fish have found that cooking leads to an increase in the wet weight content of mercury in fish, most likely as a result of moisture loss during preparation (Girard et al., 2018; Perugini et al., 2016). Since we also saw a slight drop in moisture after baking, our findings corroborate these mercury-related ones.

CONCLUSION

In conclusion, the atherogenicity (AI) and thrombogenicity (TI) indexes are two important predictors of future cardiovascular problems. They are calculated based on the concentrations of various FAs in the diet, and a higher value indicates a higher risk of developing cardiovascular disease.

REFERENCES

- Akkaya, M.R. (2018). Prediction of fatty acid composition of sunflower seeds by near-infrared reflectance spectroscopy. *Journal of Food Science and Technology*, 55(6), 2318–2325. <https://doi.org/10.1007/s13197-018-3150-x>
- Alexi, N., Kogiannou, D., Oikonomopoulou, I., Kalogeropoulos, N., Byrne, D. V., & Grigorakis, K. (2019). Culinary preparation effects on lipid and sensory quality of farmed gilthead seabream (*Sparus aurata*) and meagre (*Argyrosomus regius*): An inter-species comparison. *Food Chemistry*, 301, 125263. <https://doi.org/10.1016/j.foodchem.2019.125263>
- Bilgin, Ş., İzci, L., Günlü, A., & Bolat, Y. (2010). Effects of pan frying with different oils on some of the chemical components, quality parameters and cholesterol levels of rainbow trout (*Oncorhynchus mykiss*). *African Journal of Biotechnology*, 9(39), 6573-6577.
- Bloom, N.S. (1992). On the chemical form of mercury in edible fish and marine invertebrate tissue. *Canadian Journal of Fisheries and Aquatic Sciences*, 49, 1010–1017. <https://doi.org/10.1139/f92-113>
- Burger, J., Dixon, C., Boring, S., & Gochfeld, M. (2003). Effect of deep frying fish on risk from mercury. *Journal of Toxicology Environmental Health* Part A, 66, 817-828. <https://doi.org/10.1080/15287390306382>
- Chen, J.P., & Liu, H.B. (2020). Nutritional indices for assessing fatty acids: a mini-review. *International Journal of Molecular Sciences*, 21, 1, 5695. <https://doi.org/10.3390/ijms21165695>
- Dahl, L., Duinker, A., Næss, S., Markhus, M. W., Nerhus, I., Midtbø, L. K., & Kjellekvold, M. (2020). Iodine and mercury content in raw, boiled, pan-fried, and oven-baked Atlantic cod (*Gadus morhua*). *Foods*, 9(11), 1652. <https://doi.org/10.3390/foods9111652>
- Demirtaş Erol, N., Erdem, O.A., Yılmaz, S.T., Kayalar, H., & Cakli, S. (2022). Effects of the BHA and basil essential oil on nutritional, chemical, and sensory characteristics of sunflower oil and sardine (*Sardina pilchardus*) filets during repeated deep-frying. *LWT-Food Science and Technology*, 163, 113557. <https://doi.org/10.1016/j.lwt.2022.113557>
- EFSA (2012a). EFSA panel on dietetic products, nutrition and allergies (NDA); scientific opinion related to the tolerable upper intake level of eicosapentaenoic acid (EPA), docosahexaenoic acid (DHA) and docosapentaenoic acid (DPA). <<http://www.efsa.europa.eu/en/efsajournal/doc/2815.pdf>> Accessed 07.07.2021.

On the other hand, the hypocholesterolaemic / hypercholesterolaemic (HH) index in fish fatty acids measures the effect of fish consumption on cholesterol levels in the body. A higher HH index indicates a more hypocholesterolemic effect, which may have potential health benefits. Overall, these indices provide essential information about the health effects of different types of dietary fatty acids and can be useful for developing personalized dietary recommendations.

The n3/n6 ratio, HH, AI, and TI are the best nutritional quality indices in fish. The atherogenicity (AI) and thrombogenicity (TI) indexes were found to be lower in fried sardines. The hypocholesterolaemic/hypercholesterolaemic (HH) index of fried sardine in sunflower oil were found to be the best in terms of nutritional quality

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AUTHORSHIP CONTRIBUTION STATEMENT

Şükran Çaklı: Conceptualization, methodology; Nida Demirtaş Erol: Formal analysis, resources; Evren Burcu Şen Yılmaz: Resources, formal analysis; Pınar Baldemir: Formal analysis; Atilla Çaklı: Formal analysis.

DECLARATION OF COMPETING INTEREST

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

ETHICS APPROVAL

No specific ethical approval was necessary for this study

DATA AVAILABILITY

All relevant data is in the article

- EFSA (2012b). EFSA panel on contaminants in the food chain (CONTAM); scientific opinion on the risk for public health related to the presence of mercury and methylmercury in food. <<http://www.efsa.europa.eu/en/efsajournal/doc/2985.pdf>> Accessed 07.07.2021.
- Erdem, Ö.A., & Dinçer, M.T. (2019). Evaluation of nutritional, physical and sensory properties of flathead grey mullet's (*Mugil cephalus*) flesh after cooking. *Journal of Food Safety and Food Quality- Archiv Für Lebensmittelhygiene*, 70(5), 149-155. <https://doi.org/10.2376/0003-925X-70-149>
- Erdem, Ö.A., Alkan, B., & Dinçer, M.T. (2020). Comparison on nutritional properties of wild and cultured brown trout and Atlantic salmon. *Ege Journal of Fisheries and Aquatic Sciences*, 37(1), 37-41. <https://doi.org/10.12714/efjfas.37.1.05>
- Farag, M. M. (2013). Effect of different cooking methods on nucleic acid nitrogen bases content of fresh sardine fish and its nutritive value. *World Journal of Dairy & Food Sciences*, 8(2), 156-164. <https://doi.org/10.1016/j.foodchem.2005.08.055>
- Flaskerud, K., Bukowski, M., Golovko, M., Johnson, L., Brose, S., Ali, A., & Raatz, S. (2017). Effects of cooking techniques on fatty acid and oxylipin content of farmed rainbow trout (*Oncorhynchus mykiss*). *Food Science & Nutrition*, 5(6), 1195-1204. <https://doi.org/10.1002/fsn3.512>
- Girard, C., Charette, T., Leclerc, M., Shapiro, B. J., & Amyot, M. (2018). Cooking and co-ingested polyphenols reduce in vitro methylmercury bioaccessibility from fish and may alter exposure in humans. *Science of The Total Environment*, 616, 863-874. <https://doi.org/10.1016/j.scitotenv.2017.10.236>
- Jadán-Piedra, C., Alcántara, C., Monedero, V., Zúñiga, M., Vélez, D., & Devesa, V. (2017). The use of lactic acid bacteria to reduce mercury bioaccessibility. *Food Chemistry*, 228. <https://doi.org/10.1016/j.foodchem.2017.01.157>
- Islam, M. A., Mohibbullah, M., Suraiya, S., Sarower-E-Mahfuj, M., Ahmed, S., & Haq, M. (2020). Nutritional characterization of freshwater mud eel (*Monopterus albus*) muscle cooked by different thermal processes. *Food Science & Nutrition*, 8(11), 6247-6258. <https://doi.org/10.1002/fsn3.1920>
- IUPAC, (1979). Standard Methods for Analysis of Oils, Fats and Derivatives, 6th Edition (Fifth Edition Method II.D.19) Pergamon Press, 96-102. Oxford, UK.
- Kalogeropoulos, N., Karavoltsos, S., Sakellari, A., Avramidou, S., Dassenakis, M., & Scoullou, M. (2012). Heavy metals in raw, fried and grilled Mediterranean finfish and shellfish. *Food and Chemical Toxicology*, 50(10), 3702-3708. <https://doi.org/10.1016/j.fct.2012.07.01>
- Karimian-Khosroshahi, N., Hosseini, H., Rezaei, M., Khaksar, R., & Mahmoudzadeh, M. (2016). Effect of different cooking methods on minerals, vitamins, and nutritional quality indices of rainbow trout (*Oncorhynchus mykiss*). *International Journal of Food Properties*, 19(11), 2471-2480. <https://doi.org/10.1080/10942912.2015.1039028>
- Khansari, F. E., Ghazi-Khansari, M., & Abdollahi, M. (2005). Heavy metals content of canned tuna fish. *Food Chemistry*, 93(2), 293-296. <https://doi.org/10.1016/j.foodchem.2004.09.025>
- Kris-Etherton, P., Harris, L., & Appel, W. (2002). American Heart Association Nutrition Committee. Fish consumption, fish oil, omega-3 fatty acids, and cardiovascular disease. *Circulation*, 106, 2747-2757. <https://doi.org/10.1161/01.CIR.0000038493.65177.94>
- Larrieu, T., & Layé, S. (2018). Food for mood: Relevance of nutritional Omega-3 fatty acids for depression and anxiety. *Frontiers in Physiology*, 9, 1047. <https://doi.org/10.3389/fphys.2018.01047>
- Liao, W., Wang, G., Zhao, W., Zhang, M., Wu, Y., Liu, X., & Li, K. (2019). Change in mercury speciation in seafood after cooking and gastrointestinal digestion. *Journal of Hazardous Materials*, 375, 130-137. <https://doi.org/10.1016/j.jhazmat.2019.03.093>
- Liao, W., Zhao, W., Wu, Y., Rong, N., Liu, X., Li, K., & Wang, G. (2020). Multiple metal (loid)s bioaccessibility from cooked seafood and health risk assessment. *Environmental Geochemistry and Health*, 42(11), 4037-4050. <https://doi.org/10.1007/s10653-020-00661-9>
- Matos, J., Lourenço, H.M., Brito, P., Maulvault, A.L., Martins, L.L., & Afonso, C. (2015). Influence of bioaccessibility of total mercury, methylmercury and selenium on the risk/benefit associated to the consumption of raw and cooked blue shark (*Prionace glauca*). *Environmental Research*, 143, 123-129. <https://doi.org/10.1016/j.envres.2015.09.015>
- Morgan, J.N., Berry, M.R., & Graves, R.L. (1997). Effects of commonly used cooking practices on total mercury concentration in fish and their impact on exposure assessments. *Journal of Exposure Analysis and Environmental Epidemiology*, 7, 119-133.
- Perugini, M., Zezza, D., Tullini, S.M.R., Abete, M.C., Monaco, G., Conte, A., & Amorena, M. (2016). Effect of cooking on total mercury content in Norway lobster and European hake and public health impact. *Marine Pollution Bulletin*, 109(1), 521-525. <https://doi.org/10.1016/j.marpolbul.2016.05.010>
- Sacks, F.M., & Katan, M. (2002). Randomized clinical trials on the effects of dietary fat and carbohydrate on plasma lipoproteins and cardiovascular disease. *The American Journal of Medicine*, 113, 13-24. [https://doi.org/10.1016/S0002-9343\(01\)00987-1](https://doi.org/10.1016/S0002-9343(01)00987-1)
- Sandheinrich M., & Wiener J. (2011). Methylmercury in freshwater fish: Recent advances in assessing toxicity of environmentally relevant exposures. *Environmental Contaminations in Biota Interpreting Tissue Concentrations*, 2011(2), 169-190. <https://doi.org/10.1201/b10598-5>
- Santos-Silva, J., Bessa, R.J.B., & Santos-Silva, F. (2002) Effect of genotype, feeding system and slaughter weight on the quality of light lambs: II. fatty acid composition of meat. *Livestock Production Science*, 77, 187-194. [https://doi.org/10.1016/S0301-6226\(02\)00059-3](https://doi.org/10.1016/S0301-6226(02)00059-3)
- Schmidt, L., Bizzi, C. A., Duarte, F.A., Muller, E.J., Krupp, E., Feldmann, J., & Flores, E.M. (2015). Evaluation of Hg species after culinary treatments of fish. *Food Control*, 47, 413-419. <https://doi.org/10.1016/j.foodcont.2014.07.040>
- Ulbricht, T.L., & Southgate, D.A. (1991). Coronary heart disease: seven dietary factors. *The Lancet*, 338, Issue 8773. [https://doi.org/10.1016/0140-6736\(91\)91846-M](https://doi.org/10.1016/0140-6736(91)91846-M)
- U.S. Environmental Protection Agency (EPA) 1994. Test Methods for Evaluating Solid Waste, SW-846, 3rd ed., Office of Solid Waste and Emergency Response, Washington, D.C., November 1986; Update II, September 1994.
- Winiarska-Mieczan, A., & Grela, E.R. (2017). Content of cadmium and lead in raw, fried and baked commercial frozen fishery products consumed in Poland. *Journal of the Science of Food and Agriculture*, 97(9), 2969-2974. <https://doi.org/10.1002/jsfa.8136>
- Zhang, Y., Wang, Q., Bi, Y., Cheng, K.W., & Chen, F. (2019). Nutritional and functional activities of protein from steamed, baked, and high hydrostatic pressure treated cod (*Gadus morhua*). *Food Control*, 96, 9-15. <https://doi.org/10.1016/j.foodcont.2018.08.023>