

Impact of different cooking techniques on the fatty acid composition in phospholipid and triacylglycerol of leer (*Lichia Amia*) fillets

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SUMMARY: The fatty acid (FA) composition of leer fish (*Lichia amia*) fillets prepared using different cooking techniques was examined in this study. The phospholipid (PL) component of the fillets fried in corn and hazelnut oil contained reduced levels of PUFAs (polyunsaturated fatty acids) and SFAs (saturated fatty acids). Total PUFA, $\Sigma n6$ PUFA and linoleic acid (18:2) were detected at significantly higher levels in the triacylglycerol (TAG) fractions of fillets fried in sunflower oil and corn oil than in raw samples fried in hazelnut oil and olive oil. In comparison to raw fillets, the TAG fraction had higher rates of DHA (docosahexaenoic acid), $n3$ PUFA, $n3/n6$ in oven-baked fillets; 16:0 (palmitic acid), SFA ratios in grilling; and 18:2, PUFA, $n6$ PUFA values in microwave heating. When compared to raw fillets, it was shown that the PUFAs in the PL fraction increased when cooked in an oven or grill but did not significantly change when fried in sunflower and olive oil. In comparison to the raw fillets, $n3$ PUFAs increased in the TAG fraction using the oven and microwave techniques. Using frying methods decreased $n3$ fatty acids in both PL and TAG fractions. Cooking methods other than frying significantly altered fatty acid contents.

KEY-WORDS: Cooking methods; Leer; Phospholipid; Triacylglycerol; Vegetable oils.

RESUMEN: Impacto de diferentes técnicas de cocción en la composición de ácidos grasos en fosfolípidos y triacilglicérol de filetes de mero (*Lichia amia*). En este estudio se examinó la composición de ácidos grasos (AG) de filetes de pescado de sotavento (*Lichia amia*) preparados mediante diferentes técnicas de cocción. Los fosfolípidos (PL) de los filetes fritos en aceites de maíz y avellana contenía niveles reducidos de PUFA (ácidos grasos poliinsaturados) y SFA (ácidos grasos saturados). Se determinaron niveles significativamente más altos de PUFA totales, $\Sigma n6$ PUFA y ácido linoleico (18:2) en las fracciones de triacilglicérol (TAG) de los filetes fritos en aceite de girasol y de maíz, que en las muestras fritas en aceite de avellana y aceite de oliva. En comparación con los filetes crudos, la fracción de TAG presentó mayores niveles de DHA (ácido docosahexaenoico), $n3$ PUFA, $n3/n6$ en los filetes horneados; mayores proporciones de 16:0 (ácido palmítico) SFA, en los cocinados en parrilla; y mayores valores de 18:2, PUFA, $n6$ PUFA, en el calentamiento por microondas. En comparación con los filetes crudos, se demostró que los ácidos grasos poliinsaturados (AGPI) en la fracción PL aumentaron al cocinarlo en horno o parrilla, pero no variaron significativamente al freírlo en aceite de girasol y de oliva. En comparación con los filetes crudos, los AGPI $n3$ aumentaron en la fracción TAG al utilizar horno y microondas. El uso de métodos de fritura disminuyó los ácidos grasos $n3$ en las fracciones PL y TAG. Otros métodos de cocción, además de la fritura, alteran significativamente el contenido de ácidos grasos.

PALABRAS CLAVE: Aceites vegetales; Fosfolípidos; Leche; Métodos de cocción; Triacilglicérol.

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1. INTRODUCTION

Fish is not only an excellent source of protein from animals but also a well-known source of long-chain PUFAs, particularly DHA and EPA (eicosapentaenoic acid). Because of the high nutritional value of these FAs in the human diet, nutritionists focus on them when treating and preventing obesity (Schneedorferová *et al.*, 2015). Research shows that omega-3 fatty acids in fish oil modulate immunity and protect and improve health, which can help prevent diseases like cancer, rheumatoid arthritis, diabetes, ulcerative colitis, allergies, skin thickening, weight gain, premature birth, depression, and cardiovascular disease (Loftsson *et al.*, 2016). A lack or inadequate consumption of these fatty acids can lead to a number of problems, including memory loss, tingling in the nerves, poor vision, an increased risk of blood clots, elevated levels of triglycerides and low-density lipoprotein (LDL), and reduced function of membranes (Julius, 2006). *n3* fatty acids increase circulation, oxygen uptake, red blood cell flexibility and function by ensuring that cell walls remain elastic and flexible. AA and DHA are abundant in the phospholipids of the brain cell membrane, an organ which is rich in fatty acids. These fatty acids, after being released from the cell membrane, participate in signal transduction into various bioactive derivatives or after enzymatic transformation in the brain. It is observed that there are changes in PUFAs and the signalling pathways they regulate in neurological diseases such as Alzheimer's and major depression (Bazinet *et al.*, 2014). There's a growing interest in marine polar lipids, in particular because of their special physicochemical features and possible nutritional benefits. Due to the esterification of AA, EPA, and DHA at the sn-2 position, marine-derived PLs have characteristics that make them more nutritionally valuable, bioavailable, and absorb *n3* PUFA in the intestine more than neutral lipids (Parmentier *et al.*, 2007).

Polar lipids include phospholipids, important structural components of cell membranes, and eicosanoid precursors, which mainly include more PUFAs (De Leonardis and Macciola, 2004).

The *n3* PUFAs found in polar lipids play a role in inflammatory and allergic reactions, as well as physiological processes such as reproduction, fetal development, and birth, and antihypertensive responses. These are significant precursors to platelet-activating factors and prostaglandins (Blank *et al.*, 1992). Neutral lipids are storage lipids that often consist of monounsaturated fatty acid (MUFA) and saturated fatty acids (SFAs) (Varljen *et al.*, 2004).

TAG control and maintain the internal temperature of the body. The FA composition in fish is also influenced by the cooking techniques used. These procedures involve high temperatures that hydrolyze and oxidize nutrients.

Cooking at high temperatures is considered to make fatty acids, particularly PUFAs, particularly vulnerable to oxidation (Bastias *et al.*, 2017).

The frying techniques, lipid content in the raw fish, frying oil composition, and frying technology all play a major role in these variations (Moradi *et al.*, 2011). Cooking is a process that involves heating food for a predetermined amount of time in order to enhance its flavor and taste, increase its digestibility by altering its color, shape, and structure, deactivate harmful microorganisms and enzymes, decrease water activity, and increase its shelf life (Garcia-Arias *et al.*, 2003). Up until now, research has primarily focused on how various cooking techniques affect the overall FA contents in freshwater and marine fish (Turhan *et al.*, 2011; 2012; Başhan, 2019; Biandolino *et al.*, 2021; Biandolino *et al.*, 2023). However, there has not been much study on the FA composition of triacylglycerol, the storage lipid in fish, and structural phospholipids. The aim of this study was to investigate how various cooking techniques affect the fatty acid content of PL and TAG fractions of leer fish (*Lichia amia*) fillets by comparing them to raw fillets.

2. MATERIALS AND METHODS

2.1. Collection of samples

In the research, fish fillets were cooked with frying and non-frying techniques using leer fish (*Lichia amia*) from Karatepe Bay, and raw (uncooked) fillets were determined as standard. Each of the three fish, which weighed between 300 and 400 grams, was divided into four fillet pieces and transported to the lab in a cold chain. Every sample was thawed overnight at 4 °C in a refrigerator prior to cooking.

2.2. Cooking methods

Olive, corn, sunflower and hazelnut oil were used for frying. After the oil in the pan reached a high temperature, the fillets were fried for three minutes on each side. Each cooking process was performed three times. It was established whether fish fillets prepared without frying could sustain an interior temperature of 75 °C during the cooking process. (USDA 2023). For baking, the oven was warmed to 180 °C for 30 minutes (the center temperature was 73.6 ± 0.5 °C). After six minutes of heating in a microwave oven set to 2450 MHz and 500 W, the average temperature was 88.3 ± 1 °C. Only the steam from the water was used during the steaming process; the fillet was not in contact with any water. Steaming lasted 45 minutes at 100 °C with a central temperature of 92.5 ± 2 °C. The fillet was grilled on both sides for 8 minutes over a coal fire using the grill cooking method.

2.3. Analyses

The fillets were homogenized to evaluate the FA contents in the samples in 2:1 chloroform/methanol (Folch *et al.*, 1957). Fatty acids were then heated for two hours at 85 °C to produce methyl esters (Stanley-Samuelson and Dadd, 1983). The fractionation of total lipids in the tissue samples was achieved with the help of thin layer chromatography (TLC). The plates were air-dried and activated by keeping them in the oven at 100 °C for one hour for PL and TAG fractions.

2.4. Fatty acid analysis

FAMES were analyzed using a Shimadzu GC-2010 Plus gas chromatograph (Shimadzu Corporation, Kyoto, Japan) equipped with a flame ionization detector (FID) and a DB-23 capillary column (30 m × 0.25 mm i.d., 0.25 µm film thickness). Compressed air and hydrogen were supplied at flow rates of 400 mL/min and 30 mL/min, respectively, while helium served as the carrier gas at a flow rate of 0.5 mL/min. The injector and detector temperatures were maintained at 250 °C, and a split injection ratio of 1:20 was applied. The oven temperature was initially set at 170 °C and held for 2 min, then increased to 210 °C at a rate of 2 °C/min. Peak identification was performed by comparing retention times to those of known standards.

2.5. Statistical analysis

All analyses were conducted in triplicate, and data are presented as mean ± standard deviation (mean ± SD). Statistical evaluations were performed using SPSS version 22 (IBM Corp., Armonk, NY, USA). One-way analysis of variance (ANOVA) was employed to assess differences in fatty acid composition, followed by Tukey's post hoc test for multiple comparisons. Independent sample t-tests were used to compare fatty acid percentages, moisture contents, and total lipid levels between raw and cooked fillets. Duncan's multiple range test (1955) was also applied to evaluate differences among group means. A p-value of < 0.05 was considered statistically significant.

3. RESULTS AND DISCUSSION

3.1. The FA contents in the PL fraction with frying and non-frying cooking techniques using leer fish

Tables 1 and 2 indicate how various cooking procedures affect the FA composition of the PL fraction of leer fish. Notable differences were observed in the FA profiles between raw and cooked samples.

In the fillets fried in corn and hazelnut oils, there was a significant increase in 18:1 and Σ MUFA; while Σ SFA, DHA, Σ PUFA, n_3 PUFA, and the n_3/n_6 ratio decreased compared to the fillets fried in other oils and to the raw control.

In the raw samples, higher levels of EPA, DHA, and n_3/n_6 ratio were recorded; whereas 18:2 and Σn_6 PUFA were lower compared to the fried samples.

TABLE 1. Fatty acid composition in the phospholipid fractions of *Lichia Amia* fish fried in different vegetable oils*

Fatty acid	Raw	Sunflower oil	Olive oil	Corn oil	Hazelnut oil
C12:0**	0.38±0.03c	0.04±0.00a	0.07±0.00b	0.27±0.02c	0.06±0.00b
C14:0	0.28±0.02a	0.28±0.02a	0.31±0.02a	0.23±0.01a	0.20±0.01a
C15:0	0.20±0.01b	0.22±0.01b	0.19±0.01b	0.12±0.009a	0.13±0.01a
C16:0	26.79±2.14c	23.86±1.90b	24.48±1.95b	22.12±1.76b	19.52±1.56a
C17:0	0.06±0.00a	0.08±0.00a	0.08±0.00a	0.07±0.00a	0.06±0.00a
C18:0	10.94±0.87b	10.96±0.87b	11.35±0.90b	8.83±0.70a	7.90±0.63a
ΣSFA***	38.65±3.09d	35.44±2.83c	36.48±2.91c	31.64±2.53b	27.87±2.22a
C16:1 <i>n</i> 7	0.61±0.04b	0.37±0.02a	0.72±0.05c	0.66±0.05b	0.37±0.02a
C18:1 <i>n</i> 9	11.10±0.88a	12.66±1.01a	14.83±1.18b	29.24±2.33c	36.55±2.92d
C20:1 <i>n</i> 9	0.06±0.00b	0.06±0.00b	0.07±0.00b	0.19±0.01c	0.03±0.00a
ΣMUFA	11.77±0.94a	13.09±1.04a	15.62±1.24b	30.09±2.40c	36.95±2.95d
C18:2 <i>n</i> 6	1.82±0.14a	5.95±0.47b	4.41±0.35b	7.21±0.57c	7.57±0.60c
C18:3 <i>n</i> 6	0.28±0.02c	0.09±0.00a	0.24±0.01c	0.22±0.01c	0.15±0.01b
C18:3 <i>n</i> 3	0.18±0.01a	0.19±0.01a	0.43±0.03b	0.15±0.01a	0.20±0.01a
C20:2 <i>n</i> 6	0.16±0.01a	0.22±0.01a	0.29±0.02b	0.12±0.00a	0.15±0.01a
C20:3 <i>n</i> 6	0.06±0.00a	0.10±0.00b	0.14±0.01c	0.13±0.01c	0.08±0.00a
C20:4 <i>n</i> 6	4.43±0.35b	4.21±0.33b	4.03±0.32b	2.70±0.21a	2.31±0.18a
C20:5 <i>n</i> 3	3.65±0.29c	3.13±0.25c	2.30±0.18b	2.15±0.17b	1.73±0.13a
C22:5 <i>n</i> 3	0.91±0.07a	1.26±0.1a	2.66±0.21b	0.70±0.05a	0.70±0.05a
C22:6 <i>n</i> 3	38.11±3.04c	36.35±2.90c	33.40±2.67b	24.89±1.99a	22.29±1.78a
ΣPUFA	49.58±3.96b	51.47±4.11c	47.20±3.77b	38.27±3.06a	35.18±2.81a
Σ <i>n</i> 3 PUFA	42.83±3.42e	40.90±3.27d	38.79±3.10c	27.89±2.23b	24.92±1.99a
Σ <i>n</i> 6 PUFA	6.75±0.54a	10.57±0.84b	9.11±0.72b	10.38±0.83b	10.26±0.82b
<i>n</i> 3/ <i>n</i> 6	6.34±0.50c	3.86±0.30b	4.25±0.34b	2.68±0.21a	2.42±0.19a
ΣPUFA/ΣSFA	1.28±0.10a	1.45±0.11a	1.29±0.10a	1.20±0.10a	1.26±0.10a

*Means are the averages of 3 replicates. ** Values reported are means ± standard deviation; means followed by different letters in the same line are significantly different ($p < 0.05$) according to Tukey's test. ***SFA: saturated fatty acids, MUFA: monounsaturated fatty acids, PUFA: polyunsaturated fatty acids.

The fillets fried in corn and hazelnut oils exhibited different FA compositions from raw fish, with slight differences also noted between these two oils. Nutritional quality indices, particularly Σ*n*3 PUFA and the *n*3/*n*6 ratio, were markedly reduced in the fillets fried in vegetable oils.

The *n*3/*n*6 ratio, which was 6.34 in raw fillets, decreased to 2.42 and 4.25 in the fillets fried in hazelnut and olive oils, respectively, largely due to a reduction in DHA contents (38.11% in raw samples).

TABLE 2. Fatty acid composition in the phospholipid fractions of *Lichia Amia* cooked with different cooking methods*

Fatty acid	Raw	Oven	Grill	Microwave	Steamed
C12:0**	0.53±0.04d	0.33±0.02c	0.04±0.00a	0.14±0.01b	0.43±0.03c
C14:0	0.53±0.04c	0.37±0.02b	0.18±0.01a	0.40±0.03b	0.35±0.02b
C15:0	0.32±0.02b	0.35±0.02b	0.21±0.01a	0.40±0.03b	0.46±0.03b
C16:0	27.51±2.20a	27.19±2.17a	27.29±2.18a	29.11±2.32b	34.22±2.73c
C17:0	0.06±0.00b	0.01±0.001a	0.33±0.02c	0.07±0.00b	0.33±0.02c
C18:0	11.81±0.94a	10.77±0.86a	10.17±0.81a	13.20±1.05b	13.86±1.10b
Σ SFA***	40.76±3.26a	39.02±3.12a	38.22±3.05a	43.38±3.47b	49.65±3.97c
C16:1 <i>n</i> 7	0.74±0.05a	0.55±0.04a	0.64±0.04a	1.54±0.12b	1.15±0.09b
C18:1 <i>n</i> 9	11.69±0.93b	9.15±0.73a	9.79±0.78a	10.23±0.81b	16.27±1.30c
C20:1 <i>n</i> 9	0.12±0.00b	0.18±0.01b	0.04±0.00a	0.12±0.00b	0.31±0.02c
Σ MUFA	12.55±1.00b	9.88±0.79a	10.47±0.83a	11.89±0.95b	17.73±1.41c
C18:2 <i>n</i> 6	1.31±0.10a	1.40±0.10a	1.46±0.10a	1.90±0.15b	3.33±0.26c
C18:3 <i>n</i> 6	0.13±0.01a	0.21±0.01b	0.15±0.01a	0.12±0.01a	0.18±0.01b
C18:3 <i>n</i> 3	0.20±0.01a	0.18±0.01a	0.24±0.01a	0.33±0.02b	0.59±0.04c
C20:2 <i>n</i> 6	0.18±0.01a	0.23±0.01a	0.26±0.01a	0.18±0.01a	0.20±0.01a
C20:3 <i>n</i> 6	0.12±0.00b	0.32±0.02c	0.07±0.00a	0.08±0.00a	0.06±0.00a
C20:4 <i>n</i> 6	4.36±0.34b	3.93±0.31b	4.05±0.32b	4.32±0.34b	3.19±0.25a
C20:5 <i>n</i> 3	3.45±0.27b	3.64±0.29b	3.78±0.30b	2.65±0.21a	2.66±0.21a
C22:5 <i>n</i> 3	1.72±0.13c	1.73±0.13c	1.32±0.10c	0.97±0.07b	0.50±0.04a
C22:6 <i>n</i> 3	35.86±2.86b	39.45±3.15c	39.98±3.19c	34.14±2.73b	21.91±1.75a
Σ PUFA	47.33±2.78b	51.16±3.09c	51.31±3.10c	44.69±2.57b	32.62±2.60a
Σ <i>n</i> 3 PUFA	41.23±3.29b	45.07±3.60c	45.32±3.62c	38.09±3.04b	25.66±2.05a
Σ <i>n</i> 3PUFA	6.10±0.48a	6.09±0.48a	5.99±0.47a	6.60±0.52a	6.96±0.55a
<i>n</i> 3/ <i>n</i> 6	6.75±0.54c	7.40±0.59c	7.56±0.60c	5.77±0.46b	3.68±0.29a
Σ PUFA/ Σ SFA	1.16±0.09b	1.31±0.10b	1.34±0.10b	1.03±0.08b	0.65±0.05a

*Means are the averages of 3 replicates. ** Values reported are means $\pm$ standard deviation; means followed by different letters in the same line are significantly different ($p < 0.05$) according to Tukey's test. ***SFA: saturated fatty acids, MUFA: monounsaturated fatty acids, PUFA: polyunsaturated fatty acids.

There was limited data on the impact of the frying process on the composition of PL in fish tissues (Boselli *et al.*, 2012; Akgül *et al.*, 2024).

The Σ PUFA/ Σ SFA ratio remained above 1 in all samples, with the highest values observed for sunflower (1.45) and olive oil (1.29) fried fillets. These findings are consistent with previous reports on *Coryphaena hippurus*, where 16:0, 18:1, and DHA were predominant in the PL fraction (Akgül *et al.*, 2024).

Similarly, hazelnut oil-fried *C. hippurus* fillets demonstrated elevated 18:1 and Σ MUFA levels, while sunflower and corn oil-fried samples showed higher 18:2, Σ PUFA, and $\Sigma n6$ PUFA contents. Reductions in 16:0 and Σ SFA were also reported for hazelnut oil-fried *C. hippurus* (Akgül *et al.*, 2024), which is in agreement with the present findings. In both studies, olive oil-fried samples exhibited the highest $n3/n6$ ratios among the fried fillets.

The observed decline in long-chain $n3$ PUFAs (EPA and DHA) and the $n3/n6$ ratio in fried fillets is likely attributable to the absence of these FAs in the frying oils, as previously reported by Akgül and Başhan (2023). Farabegoli *et al.* (2019) further noted that polar and neutral lipid fractions in fish respond differently to thermal processing, with phospholipids being more resistant to oxidation than triacylglycerols. Nevertheless, the loss of water during frying may lead to apparent increases in certain FA concentrations, while highly unsaturated $n3$ PUFAs remain particularly susceptible to oxidation.

Table 2 shows the FA composition of the PL fractions in raw fillets and fillets cooked using non-frying techniques.

Raw and non-fried samples exhibited higher levels of 16:0, 18:1, and DHA. Notably, steaming resulted in elevated 16:0, Σ SFA, 18:1, and Σ MUFA levels compared to raw and other cooking methods, while DHA, Σ PUFA, $\Sigma n3$ PUFA, and the $n3/n6$ ratio were lower. The $n3/n6$ ratio was recorded as 6.75 in raw fillets, 7.40 in oven-cooked, 7.56 in grilled, 5.77 in microwave-cooked, and 3.68 in steamed fillets, with the lowest ratio observed in the steaming method.

These findings align with prior research on *C. hippurus*, where oven-cooked samples maintained higher DHA levels, and grilling resulted in the highest 18:1 content (Akgül *et al.*, 2024). Similarly, in this study, oven and grill methods produced comparable FA profiles, while microwave and steaming caused more pronounced alterations. Farabegoli *et al.* (2019) also reported minimal changes in the PL fatty acid composition of oven-baked fish species such as anchovy, sardine, and sprat. However, slight increases in 16:0, Σ SFA, EPA, DHA, Σ PUFA, and $\Sigma n3$ PUFA were noted in oven-cooked horse mackerel.

Overall, the phospholipid FA composition of leer fish fillets was more stable during oven and grill cooking compared to frying and steaming, highlighting the importance of cooking method selection in preserving the nutritional quality of fish lipids.

3.2. The FA contents in the TAG fraction with frying and non-frying techniques using leer fish

Tables 3 and 4 summarize the effects of different cooking methods on the FA composition of the TAG fraction in leer fish fillets. In the raw samples, the proportions of 16:0, 18:0, Σ SFA, 20:4, 20:5, 22:5, 22:6, and the $n3/n6$ ratio were significantly higher than in the fillets fried in vegetable oils. Conversely, 18:1 and Σ MUFA were lower in raw samples compared to the fillets fried in sunflower, olive, and hazelnut oils.

The fillets fried in sunflower and corn oils exhibited substantially elevated levels of 18:2 and $\Sigma n6$ PUFA compared to other oils (Table 3). These findings are consistent with previous studies, which reported significant increases in 18:2 and $\Sigma n6$ PUFA in fish fried in sunflower and corn oils, while 18:1 and Σ MUFA levels rose in the samples fried in olive and hazelnut oils (Akgül and Başhan, 2023). This suggests that fatty acids from frying oils are preferentially incorporated into the TAG fraction of fish muscle rather than the phospholipid fraction.

The $n3/n6$ ratio, which ranged from 0.01 to 0.17 in the fried samples, was markedly higher in raw fillets (2.43). The PUFA/SFA ratio also increased in samples fried in sunflower and corn oils. Similar to previous findings in *C. hippurus* (Akgül *et al.*, 2024), the highest proportion of 16:0 was detected in raw fish, while olive oil-fried fillets exhibited the highest Σ MUFA content. DHA remained the highest in the raw samples, although its levels were the lowest in hazelnut oil-fried *C. hippurus* and in sunflower oil-fried leer fish in the present study.

The TAG fractions of leer fish fillets cooked using non-frying methods are presented in Table 4. Certain FAs and FA groups were more prominent depending on the cooking technique. For example, grilling resulted in higher 16:0 and Σ SFA levels, while steaming led to increased 18:1 and Σ MUFA. Microwave cooking yielded higher proportions of 18:2, Σ PUFA, and $\Sigma n6$ PUFA, and oven and microwave methods preserved higher levels of DHA and $\Sigma n3$ PUFA.

TABLE 3. Fatty acid composition in the triacylglycerol fractions of *Lichia Amia* fried in different vegetable oils*

Fatty acid	Raw	Sunflower oil	Olive oil	Corn oil	Hazelnut oil
C12:0**	1.51±0.12c	0.01±0.00a	0.15±0.01b	0.10±0.01b	0.01±0.00a
C14:0	1.90±0.15c	0.14±0.01b	0.09±0.00a	0.13±0.01b	0.16±0.01b
C15:0	0.70±0.05d	0.00±0.00a	0.04±0.00b	0.05±0.00b	0.07±0.00c
C16:0	33.65±2.69d	7.74±0.61a	15.05±1.20c	11.41±0.91b	10.42±0.83b
C17:0	0.26±0.02c	0.02±0.02a	0.24±0.02c	0.06±0.00b	0.05±0.00b
C18:0	10.81±0.86c	1.98±0.15a	3.24±0.25b	2.05±0.16a	3.36±0.26b
Σ SFA***	48.83±3.90d	9.89±0.79a	18.81±1.50c	13.80±1.10b	14.07±1.12b
C16:1 n7	2.96±0.23c	0.22±0.01a	1.24±0.09b	0.23±0.01a	0.24±0.01a
C18:1 n9	23.49±1.87a	35.15±2.81b	66.10±5.28d	24.18±1.93a	58.37±4.66c
C20:1 n9	0.34±0.02e	0.01±0.00a	0.02±0.00b	0.06±0.00c	0.09±0.00d
Σ MUFA	26.79±2.14a	35.38±2.81b	67.36±5.38d	24.47±1.95a	58.70±4.69c
C18:2 n6	4.60±0.36a	53.79±4.30d	11.33±0.90b	58.69±4.69e	24.73±1.97c
C18:3 n6	0.44±0.03c	0.12±0.00a	0.25±0.02b	1.37±0.10d	0.44±0.03c
C18:3 n3	0.45±0.03c	0.11±0.00a	0.62±0.04c	0.12±0.00a	0.22±0.01b
C20:2 n6	0.26±0.02e	0.00±0.00a	0.01±0.00b	0.05±0.00d	0.03±0.00c
C20:3 n6	0.04±0.00c	0.00±0.00a	0.01±0.00b	0.01±0.00b	0.05±0.00c
C20:4 n6	1.75±0.14d	0.31±0.02c	0.14±0.01a	0.17±0.01a	0.20±0.01b
C20:5 n3	2.07±0.16d	0.06±0.00a	0.17±0.01b	0.20±0.01b	0.25±0.02c
C22:5 n3	0.95±0.07d	0.02±0.00a	0.07±0.00b	0.10±0.00c	0.08±0.00b
C22:6 n3	13.82±1.10d	0.33±0.02a	1.23±0.09c	1.02±0.08b	1.23±0.09c
Σ PUFA	24.38±1.95b	54.73±4.37c	13.83±1.10a	61.73±4.93d	27.23±2.17b
Σ n3 PUFA	17.29±1.38d	0.51±0.04a	2.09±0.16c	1.44±0.11b	1.78±0.14b
Σ n6 PUFA	7.09±0.56a	54.22±4.33d	11.74±0.93b	60.29±4.82e	25.45±2.03c
n3/n6	2.43±0.19e	0.00±0.00a	0.17±0.01c	0.02±0.00d	0.06±0.00c
Σ PUFA/ Σ SFA	0.49±0.03a	5.53±0.44c	0.73±0.05a	4.47±0.35c	1.93±0.15b

*Means are the averages of 3 replicates. ** Values reported are means $\pm$ standard deviation; means followed by different letters in the same line are significantly different ($p < 0.05$) according to Tukey's test. ***SFA: saturated fatty acids, MUFA: monounsaturated fatty acids, PUFA: polyunsaturated fatty acids.

TABLE 4. Fatty acid composition in the triacylglycerol fractions of *Lichia Amia* cooked with different cooking methods*

Fatty acid	Raw	Oven	Grill	Microwave	Steamed
C12:0**	1.51±0.12a	1.77±0.14a	1.48±0.11a	1.81±0.14a	5.92±0.47b
C14:0	1.90±0.15b	1.92±0.15b	0.90±0.07a	0.72±0.05a	2.12±0.16b
C15:0	0.70±0.05c	0.57±0.04b	0.38±0.03a	0.45±0.03a	0.29±0.02a
C16:0	33.65±2.69c	30.05±2.40b	40.50±3.24d	26.66±2.13a	30.06±2.40b
C17:0	0.26±0.02b	0.14±0.01a	0.21±0.01b	0.48±0.03c	0.68±0.05d
C18:0	10.81±0.86a	11.73±0.93a	12.16±0.97a	10.39±0.83a	10.93±0.87a
ΣSFA***	48.83±3.90b	46.18±3.69b	55.63±4.45d	40.51±3.24a	50.00±4.00c
C16:1 <i>n</i> 7	2.96±0.23d	1.99±0.15c	0.65±0.05a	1.17±0.09b	2.29±0.18c
C18:1 <i>n</i> 9	23.49±1.87c	20.16±1.61b	18.97±1.51a	21.52±1.72b	27.65±2.12d
C20:1 <i>n</i> 9	0.34±0.02b	0.46±0.03b	0.12±0.00a	0.13±0.00a	0.11±0.00a
ΣMUFA	26.79±2.14b	22.61±1.80a	19.74±1.57a	22.82±1.82a	30.05±2.40c
C18:2 <i>n</i> 6	4.60±0.36b	2.91±0.23a	4.34±0.34b	11.86±0.94c	4.85±0.38b
C18:3 <i>n</i> 6	0.44±0.03b	0.44±0.03b	0.36±0.02b	0.23±0.01a	0.39±0.03b
C18:3 <i>n</i> 3	0.45±0.03b	0.52±0.04b	0.39±0.03b	0.23±0.01a	0.57±0.04b
C20:2 <i>n</i> 6	0.26±0.02b	0.29±0.02b	0.33±0.02c	0.10±0.00a	0.21±0.01b
C20:3 <i>n</i> 6	0.04±0.00a	0.07±0.00b	0.03±0.00a	0.13±0.01d	0.10±0.01c
C20:4 <i>n</i> 6	1.75±0.14b	2.11±0.16c	2.32±0.18c	2.46±0.19c	1.19±0.09a
C20:5 <i>n</i> 3	2.07±0.16b	2.32±0.18b	3.50±0.28c	1.96±0.15b	1.42±0.11a
C22:5 <i>n</i> 3	0.95±0.07a	1.48±0.11b	0.74±0.05a	0.63±0.05a	0.74±0.05a
C22:6 <i>n</i> 3	13.82±1.10b	21.07±1.68c	12.62±1.00b	19.07±1.52c	10.48±0.83a
ΣPUFA	24.38±1.95b	31.21±2.49c	24.63±1.97b	36.67±2.93d	19.95±1.59a
Σ <i>n</i> 3 PUFA	17.29±1.38b	25.35±2.02d	17.25±1.38b	21.89±1.75c	13.21±1.05a
Σ <i>n</i> 6 PUFA	7.09±0.56b	5.82±0.46a	7.38±0.59b	14.78±1.18c	6.74±0.53a
<i>n</i> 3/ <i>n</i> 6	2.43±0.19c	4.35±0.34d	2.33±0.18c	1.48±0.11a	1.95±0.15b
ΣPUFA/ΣSFA	0.49±0.03a	0.67±0.05b	0.44±0.03a	0.90±0.07c	0.39±0.04a

*Means are the averages of 3 replicates. ** Values reported are means ± standard deviation; means followed by different letters in the same line are significantly different ($p<0.05$) according to Tukey's test. ***SFA: saturated fatty acids, MUFA: monounsaturated fatty acids, PUFA: polyunsaturated fatty acids.

Notably, the levels of EPA, DHA, and Σ*n*3 PUFA were significantly reduced in the steamed samples compared to raw and other cooking methods. The PUFA/SFA ratio was the highest in microwave-cooked fillets, and the *n*3/*n*6 ratio was the greatest in oven-cooked samples. In contrast, steaming and microwave cooking reduced the *n*3/*n*6 ratio compared to the raw fillets. These findings are in line with previous research on *C. hippurus*, where microwave cooking resulted in higher ΣPUFA levels and oven cooking preserved DHA content (Akgül *et al.*, 2024). While 16:0 was the most abundant in oven-

cooked *C. hippurus*, it was the highest in grilled leer fish in this study. Differences in the SFA and PUFA levels between studies may reflect species-specific responses to cooking and variations in tissue lipid distribution. Overall, oven and microwave cooking better preserved long-chain *n*3 PUFAs than frying or steaming.

4. CONCLUSIONS

The findings clearly demonstrate that all the cooking methods examined influenced the FA profile of leer fish fillets. Although frying had the most pronounced effect, alternative cooking methods, such as oven baking, grilling, steaming, and microwave cooking were comparatively more favorable. These techniques were associated with lower levels of less desirable FAs, such as *n*6 PUFAs, which are already abundant in the Western diet.

Notably, different vegetable oils used for frying significantly altered the FA composition of the TAG fraction. Oils rich in 18:1 and 18:2 transferred these FAs into the fish fillets, leading to their accumulation predominantly in the TAG fraction rather than in phospholipids. This finding underscores the differential partitioning of dietary FAs into lipid classes during thermal processing.

In particular, fillets fried in corn and hazelnut oils exhibited marked alterations in their FA profiles, with substantial differences in the phospholipid fractions compared to raw samples. Among the cooking methods, frying exerted the greatest impact on FA composition, while non-frying techniques preserved a higher proportion of nutritionally valuable long-chain *n*3 PUFAs.

AUTHORSHIP CONTRIBUTION STATEMENT

S. Kaçar: Writing – original draft, Writing – review & editing

M. Başhan: Investigation, Methodology, Project administration

N. Akgül: Formal analysis, Funding acquisition, Writing, Investigation, Methodology

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CONFLICT OF INTEREST STATEMENT

No potential conflict of interest was reported by the author(s).

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