

EFFECTS OF THERMAL PROCESSING ON FATTY ACIDS PROFILE OF WILD RED MULLET (*MULLUS SURMULETUS*) FROM THE MEDITERRANEAN SEA COAST OF SYRIA

Rahaf Akel^{1*}, Anwar Al-haj Ali², Moufid Yassin¹

¹Tishreen University, Latakia, Syria (Department of Analytical and Food Chemistry, Faculty of Pharmacy).

²Damascus University, Damascus, Syria (Department of Food Sciences, Faculty of Agricultural Engineering).

Received date: 03 September 2020

Revised date: 24 September 2020

Accepted date: 14 October 2020

*Corresponding author: Rahaf Akel

Tishreen University, Latakia, Syria (Department of Analytical and Food Chemistry, Faculty of Pharmacy).

ABSTRACT

Lipid, moisture and fatty acids profile of wild Red Mullet (*Mullus Surmuletus*) from Mediterranean Sea (Baniyas City - Syria) were investigated before and after (deep frying in Sunflower oil, grilling, roasting) to detect the effects of different thermal treatments on fatty acids profile especially Docosahexaenoic acid (DHA) and Eicosapentaenoic acid (EPA). samples were collected in autumn of 2019 – 2020 and analyzed by Gas Chromatography GC-FID. The results revealed significant differences between control and all heat treatments in both EPA and DHA levels (p.value < 0.05). The average of EPA and DHA estimated by g/100g of fatty acids methyl esters in control samples were 5.80 and 17.88, respectively. Frying process revealed a remarkable decrease in these two fatty acids, as they were 0.63 and 0.59, respectively. The percentage of EPA and DHA in grilling were 10.12 and 6.43 while roasting came up with levels of 11.22 and 5.82 g/ 100g of fatty acids methyl esters, respectively. Both processes were accompanied with a significant decrease in the percentage of DHA, and with a significant increase of EPA compared with the control samples. However, no significant differences were noticed in DHA percentage between grilled and roasted samples. frying was accompanied by a significant increase in the percentage of C18:2n-6 fatty acid, reaching 59.89 compared with 1.92 for the control samples which lead to very high n6/n3 ratio equal to 45.88. However, grilling and roasting came up with 0.36 n6/n3 ratio which prove their higher nutritional value comparing with frying.

KEYWORDS: Thermal processing, Red Mullet (*Mullus Surmuletus*), Fatty acids profile, GC –FID, Docosahexaenoic acid (DHA), Eicosapentaenoic acid (EPA).

INTRODUCTION

Syrian Arab Republic has a coastline of 183 km on the Mediterranean Sea. However, researches about local fish and how thermal treatments affect their basic benefits are limited. Although fish are considered to be the main source of long chain omega 3 PUFA (EPA and DHA) in Syrian diet, the average per capita consumption of fish was estimated to be about 1.5 kg in 2016 in Syria according to the Food and Agriculture Organization 2019.^[1]

Most fatty acids can be synthesized in the body. However, there is a lack in the required enzymes that produce both EPA and DHA in human bodies, for example alpha linolenic acid (ALA) can be converted into EPA and then to DHA, the conversion occurs primarily in the liver. However, the conversion process is very limited, with reported rates of less than 15%.^[2,3]

Because of that, EPA and DHA fatty acids must be available in sufficient amount in human diet.^[4]

WHO recommended regular (1-2 servings per week) of fish consumption. The serving should provide an equivalent of 200-500 mg of eicosapentaenoic and docosahexaenoic acid.^[5]

A very high omega-6/omega-3 ratio, promotes the pathogenesis of many diseases, including cancer, cardiovascular disease, inflammatory and autoimmune diseases, whereas increased levels of omega-3 PUFA (a low omega-6/omega-3 ratio) exert suppressive effects.^[6] However, improving n6/ n3 ratio should be done by increasing the n3 PUFA intake and not by decreasing n6 PUFA.^[7] Therefore, it is recommended to consume fish with high n3 PUFA, particularly EPA and DHA for the prevention of pathologies that were mentioned above.^[8]

Past studies indicate that human beings evolved on a diet with a n-6/n-3 ratio of approximately 1:1. However, some authors suggested that a ratio of n6/n3 PUFA between 1:1 and 5:1 is supposedly beneficial to health.^[6,9]

Red Mullet (*Mullus surmuletus*) considered to be one of the most available and demanded fish in Syrian market, it is also exported to Lebanon. However, it has high prices in comparison with other species and no studies are found about its fatty acids profile. Deep frying was noticed to be the first choice between the most common heat treatments for most of fish species in Syrian kitchens, followed by both grilling and roasting. studies shows a highly susceptibility to oxidation in marine lipids, these changes were suspected to belong to the high content of long- chain PUFAs in fish which are featured by their higher degree of unsaturation. that make PUFAs more sensitive to high temperature because of their lower melting points.^[10] at the same time, findings from previous studies showed that cooking losses vary greatly with fish species and cooking method.^[10] In 2014 Castro-González *et al.*, noticed that two different fish species behaved very differently when submitted to the same cooking techniques, these differences were explained by composition of raw fish, temperature, size, exposed surface and degree of postmortem ageing prior to cooking.^[8] In this article we will show fatty acids profile of the local Red Mullet (*Mullus surmuletus*) and from a nutritional point of view, we will recommend the best thermal treatments between (deep frying in Sunflower oil, grilling, roasting).

MATERIALS AND METHODS

1. Chemicals and reagents

Analytical and chromatography grade of chemicals and reagents were used in this study. Chloroform and methanol were purchased from Surechem products - England, n-Hexane from Sigma Aldrich - Germany. Potassium Hydroxide from BDH Limited Poole - England, anhydrous sodium sulphate was from Alpha Chemika - India, and (Sinopure Fish Oil – 10/70EE) from Vita Pharmaceutical - China.

2. Sample preparation

total of 5 kg of local Red Mullet (*Mullus surmuletus*) with an average weight of 54 – 110 g for each fish were purchased from Baniyas coastal city market in autumn of 2019 – 2020. Each Fish was preserved in ice and water until it reached the Food Science Department of faculty of Agriculture Engineering at Damascus University and faculty of pharmacy at Tishreen University, and was kept at -20 ° C until analysis. The samples were eviscerated then were processed without adding salt or any kind of spices in all processes.

Each fish was beheaded and chopped. the meat, muscle and skin of the fish were isolated from each fish separately, then they were minced and mixed until homogeneity using a laboratory blender.

Raw Red Mullet fishes were prepared according to the previous description and named as control samples. to prepare fried samples deep frying was performed in sunflower oil with an oil temperature (150 - 170) ° C, and a period of 3 - 5 minutes until fully cooked.^[11] Sunflower oil was used because it is the most common and demanded oil in Syrian markets.

Roasted samples were prepared using a metal bowl in a gas oven, for about 10 - 15 minutes, at a temperature of 160 ° C. However, grilled samples were prepared using a metal barbecue on charcoal for about 10 -12 minutes.

3. Moisture determination

Moisture content was determined according to (AOAC, 2000).^[12]

4. Total lipid extraction

The total lipid content of fish was determined according to Bligh and Dyer (1959) method as modified by Hanson and Olley (1963).^[13] about 10 g of fish sample was weighed into a 250-ml Erlenmeyer flask.

(8.3 – 10.3) milliliters of water (based on the known water content to give a total volume of 16 ml), 20 ml of chloroform and 40 ml of methanol were added. The mixture was homogenized for 1 min while being held in ice/water. A further 20 ml chloroform was added and then the mixture was again homogenized for 30 s. After that, 20 ml of water was added and the mixture was homogenized for a further 30 s. The homogenate was transferred into four 50-ml centrifuge tubes and centrifuged for 20 min at 2000 rpm.^[14] after centrifugation we filtered all tubes using filter papers (ZELPA 11.0 cm/ 13.0 cm) into 250 ml graduated cylinder, we used additional 30 ml of chloroform to wash the residue and centrifugation tubes,^[13] then aqueous layers were removed and 20 ml of chloroform layer containing lipid was pipetted into a dried weighed flask. The solvent was initially evaporated using a steam bath and finally in an oven at 105°C for 30 min. three replicates were applied for each sample.^[14]

5. Fatty acid methyl esters preparation

Preparation of fatty acid methyl esters (FAME) was done according to Ichihara *et al.*'s method, 1996 with minor modification. Methyl esters were prepared by trans-methylation using 2 M KOH in methanol and n-hexane. Twenty milligrams of extracted oil were dissolved in 2 ml n-Hexane followed by 4 ml of 2 M methanolic KOH. The tube was then vortexed for 2 min at room temperature then n-hexane layer was taken for GC analyses.^[14,15]

6. Gas chromatography analysis

Fatty acid profile of fish fat samples were determined by Gas Chromatograph with flam ionized detector Shimadzu GC -17AFW Model 1999 with a Split / Splitless Injection System with a glass insert, an FID ionized flame detector, a hydrogen generating device

(Shimadzu-OPGU-2200S), an air pump, a Nitrogen generator (carrier phase) (PEAK-series 600A), and computer with data output program named CLASS-GC10. A Spanish-origin Teknokroma capillary column bearing the code TR-140533 and the serial number M2056295 was used in the analysis. The column long is 30 meters with a diameter of 0.25mm, coated with a fixed phase of TRB-WAX type. The oven temperature, according to the programmed thermal system, was raised to 70 degrees Celsius at a rate of 1 ° C / minute (1C / min) for 35 minutes. The temperature of the injector and detector was set at 250 ° C and 260 ° C, respectively. The volume of the injected sample was 1 µl while the nitrogen carrier gas was at a flow of 1.0, and the split rate was 1:50. Three replicates were applied for each sample and the fatty acids were identified by comparing with the retention time of 37 components FAME mix 47 885-U (Supelco, Germany) & (Sinopure Fish Oil - 10 / 70EE) of Chinese origin, imported for Vita Pharmaceutical – Syria.

7. Statistical analysis

Statistical analysis was performed using SPSS-25 and expressed as mean±SD. One-way ANOVA, in addition to the Fisher statistic (F) were used to compare statistical significance of mean concentrations of DHA and EPA of the selected fish samples among different cooking methods at p. value < 0.05. Tukey and Games-Howell tests were performed according to the homogeneity of variances.

RESULTS AND DISCUSSION

1. Lipid and Moisture Content of Red Mullet (*Mullus surmuletus*)

The lipid and moisture content are shown in Table 1. The lipid content on the base of dry matter of control, fried in

sunflower oil, grilled and roasted samples autumn of 2019 – 2020 (October) were 3.01, 28.45, 21.04 and 21.99. However, moisture content were about 76.60, 56.63, 72.30, and 69.38, respectively. Moisture content showed significant difference between all sample treatments, the lowest moisture content noticed in fried samples followed by both roasted and grilled samples while control samples had the highest moisture content.^{[16] [17]} There were significant differences in lipid content between different processed samples and control (P<0.05). Deep fried samples had the highest lipid content which was belonged to the absorption of culinary fat and moisture loss as a result of thermal processing, in 2006 Nomikos *et al*, showed that Frying resulted in an increase of the levels of total lipids in six different species of fresh fish, namely rainbow trout (*Oncorhynchus mykiss*), golden trout (*Onchorhynchus aguabonita*), sea bass (*Dicentrarchus labrax*), haddock (*Melanogrammus aeglefinus*), coley (*Pollachius virens*) and plaice (*pleuronectes platessus*).^[18] No significant difference was noticed in the lipid content of grilled and roasted samples, but they were significantly higher than control samples, this increase in lipid content belonged to moisture loss while thermal processing. the same results were noticed in (*Rhamdia quelen*), whereas an increase of lipid and protein content in roasted samples was noticed and explained by moisture loss.^[19] Another study on (*Scomberomorus guttatus*) was shown an increase of lipid content of grilled samples comparing with control and this increase was attributed to the same reason.^[20]

Table 1: Comparative analysis between moisture and total lipids on the base of dry matter.

Component	Control	Fried	Grilled	Roasted
Lipid	3.01 ± 0.13 ^a	28.45 ± 0.82 ^b	21.04 ± 0.25 ^c	21.99 ± 0.49 ^c
Moisture	76.60 ± 0.39 ^a	56.63 ± 0.76 ^b	72.30 ± 0.43 ^c	69.38 ± 0.12 ^d

(a-b-c-d) means in a row with the identical letters are not significantly different (P<0.05). Values were presented as mean ± SD (n=3)

2. Fatty acids (FA) Composition

Fatty acids profile of wild Red Mullet (*Mullus surmuletus*) from the Mediterranean Sea - Baniyas city coast before and after thermal processing are shown in Table 2, levels of fatty acids were estimated by g/100g of fatty acids methyl esters. A total of 22 fatty acid were identified in control samples. The composition of saturated (SFA), monounsaturated (MUFA), and polyunsaturated (PUFA) were found to be 41.57, 22.66, 35.77 g/100g, respectively. while, n-6/n-3 ratio was about 0.38. SFA levels were noticed to be the highest between other groups in local red mullet. In this study results were quite differ from Öksüz *et al*. (2010) who reported that striped red mullet SFA, MUFA and PUFA in march of 2008 were about 36.72, 41.83, and 18.92,

respectively.^[21] However, previous studies declared multiple reasons that can affect fatty acids composition between & within fish species depending on the geographical location of fishing ground, season, food availability, age, size, the maturation status of fishes, PH, salinity and temperature of water.^[9,16,21] Polat *et al*. (2009) showed the difference in moisture, lipid, and fatty acids contents of red mullet (*Mullus barbatus*) captured from Iskenderun bay, northeastern Mediterranean in autumn, winter and spring and declared that the fatty acid contents of red mullet were influenced by seasonality.^[22] Moradi *et al*. (2011) showed that saturated fatty acids of fish lipids (SFA) are dominated by palmitic acid (C16:0), followed by myristic acid (C14:0) and stearic acid (C18:0).^[16,23] However,

monounsaturated fatty acids (MUFA) are dominated by oleic acid C18:1(n-9), followed by the three times less abundant palmitoleic acid C16:1(n-7). At the same time, they noticed that in aquatic animals, polyunsaturated fatty acids (PUFA) are usually long-chain and quantitatively are mostly EPA C20:5(n-3) and DHA C22:6 (n-3). However, in most carnivorous fish and invertebrates, DHA is usually more abundant than EPA; up to 2–3 times the proportion of EPA, and the proportion of EPA is higher in shellfish (crustaceans, cephalopods, and bivalves) than in finfish (bony fish, sharks, and rays).^[23,24] The same proportion was noticed in this study, Palmitic Acid (C16:0) was the most abundant saturated fatty acid in control samples followed by stearic acid C18:0 and myristic acid C14:0 with an average of 24.27, 7.32 and 3.60 g/100g of fatty acids methyl esters. At the same time, DHA (C22:6 n-3), and oleic acid (C18:1n-9) were the most abundant between PUFA and MUFA with an average of 17.88 and 12.05

g/100g of fatty acids methyl esters, respectively. However, The average of EPA was about 5.80 g/100g.^[16]

3. Effects of thermal processing on fatty acids profile

Total of 18 fatty acids were detected in deep fat fried samples, the results showed an absence of C17:1, C20:2n6, C20:4n-6, and C22:5n-3. However, a new peak with a level of 0.53 g/100g was appeared and wasn't identified in this study, the same new peak was noticed in sunflower oil that was used in frying process. Changes in fatty acids profile were attributed to fat exchange between the fish and culinary oil and sunflower oil absorption which resulting in modification of fatty acids profile.^[8,25,26]

Grilling and roasting showed 21 fatty acids, no additional peaks were appeared. However, both processes showed an absence of C20:2n-6.

Table 2: Fatty acid profiles (g/ 100g of total fatty acids*) of local Red Mullet (*Mullus surmuletus*).

Fatty acid %	Control	Fried	Grilled	Roasted
C14:0	3.60 ± 0.12	0.34 ± 0.08	4.51 ± 1.01	4.87 ± 0.06
C15:0	1.13 ± 0.03	0.09 ± 0.02	0.87 ± 0.16	0.85 ± 0.05
C16:0	24.27 ± 0.42	7.55 ± 0.59	26.53 ± 2.23	23.73 ± 0.34
C17:0	1.05 ± 0.05	0.09 ± 0.02	1.06 ± 0.04	0.94 ± 0.06
C18:0	7.32 ± 0.10	3.99 ± 0.11	6.69 ± 0.27	5.93 ± 0.08
C20:0	1.83 ± 0.20	0.11 ± 0.02	1.00 ± 0.04	1.15 ± 0.03
C24:0	2.38 ± 0.36	0.17 ± 0.03	2.07 ± 0.22	2.27 ± 0.15
SFA	41.57 ± 0.41 ^a	12.34 ± 0.78 ^b	42.74 ± 3.07 ^{ac}	39.74 ± 0.59 ^c
C16:1n-7	5.06 ± 0.43	0.44 ± 0.11	9.13 ± 0.45	9.80 ± 0.82
C17:1	0.55 ± 0.01	ND	0.44 ± 0.08	0.42 ± 0.04
C18:1n-9	12.05 ± 0.17 ^a	23.59 ± 1.04 ^b	16.06 ± 0.69 ^c	17.04 ± 0.42 ^c
C18:1n-7	4.39 ± 0.10	1.17 ± 0.86	7.11 ± 1.97	7.48 ± 0.30
C20:1n-9	0.62 ± 0.11	0.10 ± 0.00	0.48 ± 0.10	0.62 ± 0.05
MUFA	22.66 ± 0.33 ^a	25.30 ± 1.79 ^a	33.22 ± 1.02 ^b	35.37 ± 1.39 ^b
C18:2n-6 (LA)	1.92 ± 0.05 ^a	59.89 ± 1.06 ^b	1.26 ± 0.06 ^c	1.26 ± 0.02 ^c
C18:3n-3(ALA)	0.24 ± 0.07 ^a	0.09 ± 0.05 ^b	0.28 ± 0.04 ^a	0.24 ± 0.02 ^a
C18:4n-3(SA)	0.54 ± 0.01	0.21 ± 0.02	0.39 ± 0.03	0.39 ± 0.04
C20:2n-6	0.36 ± 0.15	ND	ND	ND
C20:3n-6	0.50 ± 0.20	0.28 ± 0.06	0.40 ± 0.10	0.49 ± 0.16
C20:4n-6	5.17 ± 0.09	ND	3.22 ± 0.13	3.01 ± 0.16
C20:5n-3 (EPA)	5.80 ± 0.08 ^a	0.63 ± 0.08 ^b	10.12 ± 0.52 ^c	11.22 ± 0.63 ^d
C22:2n-6	1.68 ± 0.20	0.15 ± 0.02	1.31 ± 0.22	1.77 ± 0.18
C22:5n-3	1.67 ± 0.05	ND	0.63 ± 0.16	0.68 ± 0.11
C22:6n-3 (DHA)	17.88 ± 0.31 ^a	0.59 ± 0.09 ^b	6.43 ± 1.30 ^c	5.82 ± 0.81 ^c
PUFA	35.77 ± 0.23 ^a	61.83 ± 1.08 ^b	24.04 ± 2.19 ^c	24.89 ± 1.96 ^c
Not Identified	ND	0.53 ± 0.10	ND	ND
n-6	9.63 ± 0.45 ^a	60.31 ± 1.10 ^b	6.20 ± 0.24 ^c	6.53 ± 0.47 ^c
n-3	25.59 ± 0.27 ^a	1.31 ± 0.01 ^b	17.46 ± 1.93 ^c	17.96 ± 1.50 ^c
n-6/n-3 ratio	0.38 ± 0.02 ^a	45.88 ± 1.26 ^b	0.36 ± 0.03 ^a	0.36 ± 0.005 ^a

*Values were presented as mean ± SD as percentages of total fatty acids, (n=3).
 **(a-b-c-d) means in a row with the identical letters are not significantly different (P<0.05).
 *** ND: Not Detected

4. Effects of thermal processing on Eicosapentaenoic acid (EPA) and Docosahexaenoic acid (DHA)

EPA levels of control, fried, grilled and roasted samples were 5.80, 0.63, 10.12 and 11.22 estimated by g/100g of fatty acids methyl esters, respectively. Significant differences were noticed between control and all heat treatments (P value < 0.05).

Fried samples lost 89 % of EPA comparing with control samples. However, grilled and roasted samples' EPA levels were noticed to be about twice as EPA levels in control samples. A significant increase was noticed in roasted samples comparing with grilling.

DHA levels of control, fried, grilled and roasted samples were 17.88, 0.59, 6.43 and 5.82 estimated by g/100g of fatty acids methyl esters, respectively. Significant differences were noticed between control samples and all heat treatments (P value < 0.05).

Fried samples lost 96.7% of DHA comparing with control samples. However, grilled and roasted showed a significant decrease in DHA and lost about 64% - 67% of DHA comparing with control. No significant difference was noticed between grilled and roasted samples.

The significant decrease of EPA and DHA in fried samples belonged to absorption of the culinary fat into the fish, moisture loss, leaching of fat soluble molecules out of fishes and oxidation reactions with free radicals generated in the hot culinary fat.^[16,27] The significant increase in EPA in grilled and roasted samples belonged to exposure to heat and moisture loss.^[8,26,28]

Deep-fat frying in sunflower oil came up with a significant increase in the level of Linoleic acid C18:2n-6 (LA) which noticed to be 1.92, 59.89, 1.26 and

1.26 g/100g of fatty acids methyl esters in control, fried, grilled and roasted samples, respectively. This higher increase belonged to the absorption of sunflower oil which noticed to have a level of C18:2 equal to 48.3 - 74.0 estimated by g/100g of fatty acids methyl ester in Syria according to the Syrian National Standard for Sunflower oil (2015), and about 62.5 g/100g in the used sunflower oil in this study.^[29]

5. Effects of thermal processing on saturated (SFA), monounsaturated (MUFA), and polyunsaturated (PUFA)

SFA, MUFA and PUFA levels in control, fried, grilled and roasted samples are shown in Fig. 1.

SFA levels were about 41.57, 12.34, 42.74 and 39.74, MUFA levels were noticed to be about 22.66, 25.30, 33.22 and 35.37. However, PUFA levels were about 35.77, 61.83, 24.04 and 24.89 in control, fried, grilled and roasted samples, respectively. No significant difference was noticed in SFA between control and grilled samples, and between grilled and roasted samples. However, a significant decrease in SFA was noticed in fried and roasted samples. MUFA came up with significant differences between control and all heat treatment except frying. Roasted samples had the highest level of MUFA followed by grilled samples with no significant difference between both processes. However, fried samples had the lowest level of MUFA between heat treatments.

PUFA came up with significant differences between control and all sample treatments. However, no significant difference was noticed between grilled and roasted samples. Frying came up with the highest PUFA levels which belonged to the higher levels of C18:2n-6 in sunflower oil as we declared above.^[29]

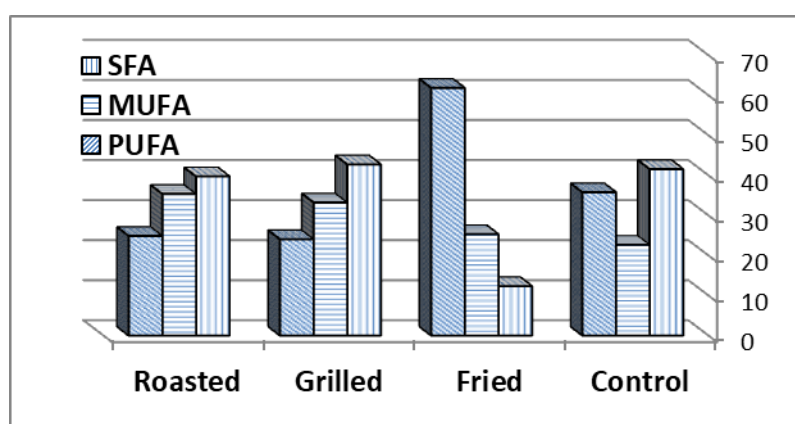


Fig. 1: SFA, MUFA and PUFA levels in control, fried, grilled and roasted samples.

6. Omega 6 (n-6), Omega 3 (n-3) fatty acids and n-6/n-3 ratio

Omega 6 (n-6), Omega 3 (n-3) and (n-6/n-3 ratio) levels in control, fried, grilled and roasted samples are shown in Fig. 2.

Omega 6 (n-6) levels were noticed to be about 9.63, 60.31, 6.20 and 6.53. At the same time, Omega 3 (n-3) levels were about 25.59, 1.31, 17.46 and 17.96 in control, fried, grilled and roasted samples, respectively. Frying came up with the highest n-6 and lowest n-3 levels and that belonged to the higher content of C18:2n-

6 which came from frying oil. However, grilling and roasting showed lower levels of n-6 and n-3 fatty acids while comparing with control and a higher n-3 levels with lower n-6 levels while comparing with fried samples. significant differences were noticed between control and all thermal treatments (P. value<0.05). However, no significant difference was noticed between grilled and roasted samples in both n-6 and n-3 fatty acids. As a result n-6/n-3 ratio in control, fried, grilled and roasted samples were notice to be 0.38, 45.88, 0.36, and 0.36, respectively. No significant differences were noticed between control, grilled and roasted samples.

However, frying illustrated the highest ratio of n-6/n-3 causing the loss of the expected nutritional value of n-3 fatty acids in Red Mullet fish. Results showed that grilling and roasting of red mullet are close in their nutritional value and have better fatty acids composition comparing with frying, whereas n-3 fatty acids are higher than n-6 fatty acids in both heat treatment. As a result, the regular consumption of grilled and roasted red mullet (*Mullus surmuletus*) fish which were noticed to have high n-3 fatty acids and low n-6 fatty acids is preferred and will contribute in healthy diet with better n-6/n-3 ratio comparing with frying.^[6]

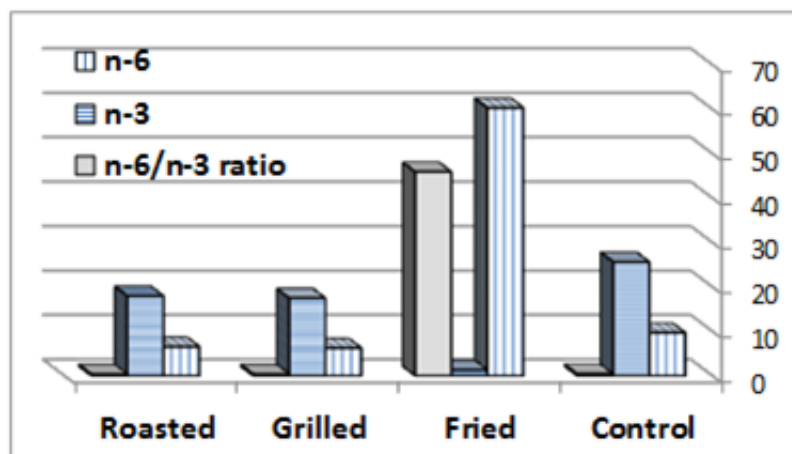


Fig. 2: n-3 and n-6 levels in control, fried, grilled and roasted samples

CONCLUSIONS

According to our study lipid and moisture content of local wild Red Mullet (*Mullus surmuletus*) in autumn of 2019 - 2020 are about 3.01, and 76.60%, with a variety of 22 different fatty acids and a level of EPA and DHA equal to 5.80 and 17.88 g/100g of fatty acids methyl esters, respectively.

Both grilling and roasting treatments were companied with a significant decrease in the percentage of DHA, and with a significant increase of EPA compared with control samples. Roasting came up with the highest levels of EPA. However, no significant differences were noticed in DHA levels between grilled and roasted samples, also both processes showed n-6 levels less than control and fried samples, and higher n-3 levels comparing with frying. Frying in sunflower oil caused a loss of the nutritional value that is expected to be gained by the consumption of Red Mullet (*Mullus surmuletus*), it companied with a significant increase of n-6 fatty acids and decrease of n-3 fatty acids with the lowest EPA and DHA levels. No significant differences were noticed in n-6/n-3 ratio between control, grilled and roasted samples which make grilling and roasting good choices for treating red mullet(*Mullus surmuletus*).

ACKNOWLEDGEMENT

We are very grateful to Food Science Department and to the National Commission For Biotechnology (NCBT) of

Syria, department of Food Biotechnology - faculty Agricultural Engineering at Damascus University and to faculty of pharmacy in Tishreen University for the facilities.

CONFLICT OF INTEREST

We declare that we have no conflict of interest.

REFERENCES

1. Fishery and Aquaculture Country Profiles. Syrian Arab Republic (2019). Country Profile Fact Sheets. In: FAO Fisheries Division [online]. Rome. Updated 1 October 2007. [Cited 28 September 2020]. <http://www.fao.org/fishery/>.
2. Coates PM, Betz JM, Blackman MR, Cragg GM, Levine M, Moss J, et al. Encyclopedia of dietary supplements: CRC Press, 2010.
3. Nesheim MC, Oria M, Yih PT, Resources N, Council NR. DIETARY RECOMMENDATIONS FOR FISH CONSUMPTION. A Framework for Assessing Effects of the Food System: National Academies Press (US), 2015.
4. Lunn J, Theobald H. The health effects of dietary unsaturated fatty acids. Nutrition Bulletin, 2006; 31(3): 178-224.
5. Kris-Etherton PM, Grieger JA, Etherton TD. Dietary reference intakes for DHA and EPA. Prostaglandins, Leukotrienes and Essential Fatty Acids, 2009; 81(2-3): 99-104.

6. Simopoulos AP. The importance of the ratio of omega-6/omega-3 essential fatty acids. *Biomedicine & pharmacotherapy*, 2002; 56(8): 365-79.
7. Deckelbaum RJ. n-6 and n-3 Fatty acids and atherosclerosis: ratios or amounts? : *Am Heart Assoc*, 2010.
8. Castro-González I, Maafs-Rodríguez AG, Romo FP-G. Effect of six different cooking techniques in the nutritional composition of two fish species previously selected as optimal for renal patient's diet. *Journal of food science and technology*, 2015; 52(7): 4196-205.
9. Strobel C, Jahreis G, Kuhnt K. Survey of n-3 and n-6 polyunsaturated fatty acids in fish and fish products. *Lipids in Health and Disease*, 2012; 11(1): 1-10.
10. Choo PY, Azlan A, Khoo HE. Cooking methods affect total fatty acid composition and retention of DHA and EPA in selected fish fillets. *SCIENCEASIA*, 2018; 44(2): 92-101.
11. Gladyshev MI, Sushchik NN, Gubanenko GA, Demirchieva SM, Kalachova GS. Effect of way of cooking on content of essential polyunsaturated fatty acids in muscle tissue of humpback salmon (*Oncorhynchus gorbuscha*). *Food Chemistry*, 2006; 96(3): 446-51.
12. Chemists AoOA, Horwitz W. Official methods of analysis: Association of Official Analytical Chemists Washington, DC, 1975.
13. Bligh E, Dyer W. Canadian journal of biochemistry and physiology. A rapid method of lipid extraction and purification, 1959; 37: 911-7.
14. Ozogul Y, Şimşek A, Ballıçlı E, Kenar M. The effects of extraction methods on the contents of fatty acids, especially EPA and DHA in marine lipids. *International journal of food sciences and nutrition*, 2012; 63(3): 326-31.
15. Ichihara Ki, Shibahara A, Yamamoto K, Nakayama T. An improved method for rapid analysis of the fatty acids of glycerolipids. *Lipids*, 1996; 31(5): 535-9.
16. Moradi Y, Bakar J, Motalebi A, Syed Muhamad S, Che Man Y. A review on fish lipid: composition and changes during cooking methods. *Journal of Aquatic Food Product Technology*, 2011; 20(4): 379-90.
17. Puwastien P, Judprasong K, Kettwan E, Vasanachitt K, Nakngamanong Y, Bhattacharjee L. Proximate composition of raw and cooked Thai freshwater and marine fish. *Journal of Food Composition and Analysis*, 1999; 12(1): 9-16.
18. Nomikos T, Karantonis HC, Skarvelis C, Demopoulos CA, Zabetakis I. Antiatherogenic properties of lipid fractions of raw and fried fish. *Food Chemistry*, 2006; 96(1): 29-35.
19. Weber J, Bochi VC, Ribeiro CP, Victório AdM, Emanuelli T. Effect of different cooking methods on the oxidation, proximate and fatty acid composition of silver catfish (*Rhamdia quelen*) fillets. *Food Chemistry*, 2008; 106(1): 140-6.
20. Bakar J, Rahimabadi EZ, Man YC. Lipid characteristics in cooked, chill-reheated fillets of Indo-Pacific king mackerel (*Scomberomorus guttatus*). *LWT-Food Science and Technology*, 2008; 41(10): 2144-50.
21. Öksüz A, Özyılmaz A, Küver Ş. Fatty acid composition and mineral content of *Upeneus moluccensis* and *Mullus surmuletus*. *Turkish Journal of Fisheries and Aquatic Sciences*, 2011; 11(1): 69-75.
22. Polat A, Kuzu S, Oezyurt G, Tokur B. Fatty acid composition of red mullet (*Mullus barbatus*): a seasonal differentiation. *Journal of Muscle Foods*, 2009; 20(1): 70-8.
23. Ackman R. *Seafood lipids. Seafoods: Chemistry, processing technology and quality*: Springer, 1994; 34-48.
24. Dunstan GA, Baillie HJ, Barrett SM, Volkman JK. Effect of diet on the lipid composition of wild and cultured abalone. *Aquaculture*, 1996; 140(1-2): 115-27.
25. Ansorena D, Guembe A, Mendizábal T, Astiasarán I. Effect of fish and oil nature on frying process and nutritional product quality. *Journal of food science*, 2010; 75(2): H62-H7.
26. Hosseini H, Mahmoudzadeh M, Rezaei M, Mahmoudzadeh L, Khaksar R, Khosroshahi NK, et al. Effect of different cooking methods on minerals, vitamins and nutritional quality indices of kutum roach (*Rutilus frisii kutum*). *Food chemistry*, 2014; 148: 86-91.
27. Little S, Armstrong S, Bergan J. Factors affecting stability and nutritive value of fatty acids: culinary practices. *Fatty acids in foods and their health implications*, 2000; (Ed. 2): 427-37.
28. Ersoy B, Özeren A. The effect of cooking methods on mineral and vitamin contents of African catfish. *Food Chemistry*, 2009; 115(2): 419-22.
29. SASMO Organization: Damascus —Syria - Ministry of Industry – Yousef al-Azmaa Sq – .Maisallon Street Tel + 963 11 2314239 - Fax + 963 11 2314239- P.O Box 11836 – Damascus https://www.unrwa.org/sites/default/files/annex_g_-_syrian_national_standard_for_sunflower_oil.pdf.