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Effect of Different Cooking Methods on Nucleic Acid Nitrogen Bases Content of Fresh Sardine Fish and Its Nutritive Value

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Abstract: Sardine fishisarich source with protein, minerals, vitamins and polyunsaturated fatty acids but also, it containshigh quantity of nucleic acid nitrogen bases (NAN) which lead to gout disease. So, different cooking methods such as; boiling, frying, grilling, roasting and by microwave were used for studying the effect them on NAN bases content of fresh Sardine fish and its nutritive value. The wet heat was the most effect on nucleic acid nitrogen (NAN) bases content of Sardine fish, the decrease percentreached to 48% after boiling method for 25min. On the contrast, the dry heat increased the NAN bases content in samples to +3% by grilling method for 15min. The nutrient composition of fresh Sardine fish such as; protein and ash increased with increasing the cooking durationwhereas, both moisture and fat content were contrast. Total Volatile Base-Nitrogen (TVN) and cooking loss reached to the highest value were 78mg/100g and 46% after grilling for 15min respectively, whereas, both Thiobarbituric acid (T.B.A) and Trimethylamine (T.M.A) recorded the highest values were 1.96mg/kg and 3.84mg/100g after frying for 15min. Most of macro-elements minerals increased after grilling for 15min followed cooking by microwave for 7min whereas, micro-elements increased after frying for 15min and roasting for 25min but all minerals decreased after boiling method for 25min except of Cu and Se decreased by frying to compare with fresh samples.Fat- soluble vitamins such as; A, D, E and K increased after frying for 15min Whereas, allvitamins decreased after boiling method for 25min compared with fresh samples. Total essential amino acids content in fresh Sardine samples was 490mg/g protein more than essential amino acids standard (FAO/WHO/UNU) was 460mg/g protein. Essential amino acids recorded the highest values after grilling method for 15min and the total reached to 523mg/g protein.Sardine fish is rich with polyunsaturated fatty acids especially ecosapentaaenoic acid (EPA), decosapentaaenoic acid (DPA) and decosahexaaenoic acid (DHA), the total of them was 12.97g/100g fatty acids and increased after all cooking methods until reached to 17.75g/100g fatty acids after frying method for 15min.

Key words: Sardine Fish · Cooking Methods · NAN · Nutritive Value

INTRODUCTION

High-purine foods are also high-protein foods and they include organ meats like kidney, fish like mackerel, herring, sardines and mussels and also yeast. Purines are metabolized into uric acid, when cells die and get recycled; the purines in their genetic material also get broken down. Uric acid is the chemical formed when purines have been broken down completely, it's normal and healthy for uric acid to be formed in the body from breakdown of purines, for example, uric acid serves as an antioxidant and helps prevent damage to our blood vessel linings, so a continual supply of uric acid is important for protecting our blood vessels. Also, in a case of severe gout, total daily purine intake must be decreasedto 100-150 milligrams. A 3.5 ounce serving of some foods, all by itselfcan contain up to 1,000 milligrams of purines. These foods include anchovies, herring, kidney, liver, mackerel, meat extracts, mincemeat, mussels, sardines and yeast [1]. According to the Gordon Research Institute, seafood and fish are potent sources of nucleic acids, Sardines contain one of the highest levels of nucleic acids, accounting for 1.5 percent of its nutritional components, while red meat, for instance, contains only 0.05 percent. Although the fundamental importance of nucleic acids in the proper functioning of cells and body

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systems has been established, people suffering from gout should avoid the excessive consumption of fish, seafood and other foods rich in nucleic acids [2].

Gout is a type of arthritis, it is caused by having too much of the chemical uric acid in your bloodstream, uric acid is the waste product created when the body breaks down purines (a type of protein found in many foods and all of your cells). Increased levels of uric acid in your blood may occur if, for example, your kidneys cannot efficiently remove it, you have a rare genetic abnormality, or because your diet and lifestyle increase the amount of uric acid that you produce. If levels of uric acid are high for prolonged periods, needle-like crystals can start to form in your tissues, resulting in swollen, painful joints. Your diet plays an important role in both causing gout and reducing the likelihood of suffering further painful attacks of gout. If you already suffer from gout, eating a diet that is rich in purines can result in a five-fold increase in gout attacks [3].

The fish obtained from sea and freshwaters are nutritionallyimportant, a great deal of seafood is a perfect vitamin and mineralsource. The high protein levels, with good digestibility and also lowfat content, are advantages of seafood, thenutritive value of fish can be affected by cooking methods [4]. Processing by heat is hypothesized to increase food digestibility due to breakdown of complex proteins and carbohydrates. Despite this, however, vitamins, minerals, some essential amino acids and other beneficial nutrients are lost [5]. The percentages of protein, fat, moisture in fresh, dried green catfish and sardine meal 18.43, 65.99, 68.80; 4.93, 22.40, 7.78 and 75.75, 7.04, 6.17, respectively. Vitamin E content in dried green catfish (264g/100g) ishigher than that in sardine meal (84g/100g) while calcium and phosphorus in dried green catfish arelower than those in sardine meal [6].

Eight cooked species of fish and one species of shrimps (grilled, curried, fried and cooked in rice) are commonly consumed in Bahrain for their proximate mineral and heavy metal content,fishes has a considerable content of sodium ranging from 120-600 mg/100g, potassium (310-560 mg/100g) phosphorous (200- 330 mg/100g), magnesium (26-54 mg/100g) and zinc (0.4-2.0 mg/ 100g), while the other minerals are present to a lower extent. Also, thestudiesrevealed that the protein content isin the range of 22.8-29.2 g/100g, while the fat content isbetween 2.9-11.9 g/100g [7].

The conventional and microwave oven cooking of rainbow trout fillets do not cause significant losses, the amount of lipid lost. Cooking significantly increase mono unsaturated fatty acids (MUFA) content of fish fillets. The results indicate a relatively high durability of n-3 poly unsaturated fatty acids (PUFA) contained in fish lipids. As a result, both conventional oven and microwave oven cooking methods can be preferred without significant loss of n-3 PUFA [8].

Fish is treated by various processes, such as boiling, grilling, baking and frying, before consumption. Heating is applied to food to enhance its flavor and taste, inactivate pathogenic microorganisms and increase shelf life. Also, cooking processes affect purine content where, the boiling high-purine foods in water can cause break-down of the purine-containing components (called nucleic acids) and eventual freeing up of the purines for absorption [9].

Sardine is one of very popular fishes in Japan, it is often eaten as dried sardine, grilled, simmered with soy sauce or miso (soy paste) sauce, Tenpura or Sashimi (only fresh one), it is a good calcium supplierif small ones (grilled or fried) are eaten wholly and is also rich in omega-3oilsand other important nutrients for a healthy pregnancy. At least 150 grams (5 ounces) of cooked fish each week must be eaten, as recommended in Canada's Food Guide [10].

Cooking of tilapia fish steakssignificantly increase the contents of eicosa-pentaenoic acid (EPA) and decosa-hexaenoic acid (DHA) in tilapia meat, no significant ($p>0.05$) impact is observed on the contents of decosa-pentaenoic acid (DPA). Therefore tilapia meat appears to be a valuable source of n-3 PUFAs for preventing coronary heart disease (CHD), Brain development and mental health, Hypertension, Diabetes, obesity, Cancer, thrombosis, lung disease and some other diseases. Also, the fatty acid composition of the cooked tilapia fish steaks is significantly decreased in the levels of monounsaturated fatty acids and increased in the n-3/n-6 Poly-Unsaturated. Also, cooking of tilapia fish steaks has a significant influence on the levels of thiobarbituric acid (TBA), salt, total volatile basic nitrogen (TVBN) and total bacterial count (TPC) of raw/fresh meat of tilapia, which are 0.77±0.01mg%, 0.58±0.01%, 3.57±0.01mg% and 4.9 x104 cfu/g change to 1.61±0.06mg%, 0.41±0.01%, 3.59±0.01mg% and <1x102 cfu/g respectively, after cooking [11].

Lowpercentage of saturated fatty acids (4.25 and 2.35%) is present insardine and tilapia, respectively, while this percentage increase in mullet fishby 6.25% of saturated fatty acids. Also, monounsaturated fatty acids percentage decrease is lowin sardine and mullet fish by 3.84 and 11.72% respectively. Polyunsaturated fatty acidsare highin all types of fish by varying percentages.

U.S. health organizations recommend a daily eicosa-pentaenoic acid (EPA) and decosa-hexaenoic acid (DHA) intake of 250 mg for most consumers and 1000 mg for people with cardiovascular disease. FAO/WHO experts recommend a daily DHA of at least 200 mg for pregnant or breastfeeding women [2].

The aim of this study was to investigate the effect of different cooking methods on nucleic acid nitrogen bases (NAN) content of Sardine fish and its nutritive value.

MATERIALS AND METHODS

Samples and Treatments: Freshly Sardine fish samples (*Sardinapilchardus*) were purchased from a local market. The fish were immediately packed in ice and transported to the laboratory, the Sardine fish samples were washed after removal of heads, viscera, tails, bone and skin, the edible portions were analyzed. Fifth cooking treatments like; boiling, frying, grilling, roasting and microwave were carried out batches. The first batch (boiled) was cooked in boiled water (1/5; w/v) at 100°C for 20 and 25 min, the second batch (fried) was cooked in deep frying oil at 180°C for 10 and 15 min, the third batch (grilled) was cooked on direct fire at 250°C for 10 and 15 min, the fourth batch (roasted) was cooked in oven at 200°C for 20 and 25 min and then, the fifthbatch was placed on a dish and cooked by microwave oven (600W, Teco. Korea) for 5 and 7min.

Analyses: Total nucleic acid nitrogen bases (NAN) of fresh and cooked Sardine fish were determined [12]. Proximate chemical analysis wasdetermined [13]. Total volatile base-nitrogen (TVB-N) of raw materials was determined by the steam distillation [14]. Thiobarbituric acid (TBA), mg malonaldehyde/kg sample) was determined by a distillation method and the absorbance was measured against the blank at 538nm using [15]. TBA number (mg malonaldehyde per kgsample) = 7.8 D. Trimethylamine (TMA) of raw materials was determined by the Modified Dyer Picrate method [16]. Minerals were analyzed by atomic absorption [17]. Vitamins were analyzed by high pressure liquid chromatography (HPLC) and micro bioassay method as described by Kamman *et al*. [18].

Amino acid composition was determined by a high speed amino acid analyzer (Hitachi Model L-8500, Japan) and tryptophan was analyzed [19]. Total lipids of the fish muscle were extracted using the method described [20]. Tissue samples weighing 3g were used for the extraction of total lipid and the fatty acids in the total lipids were converted into fatty acid methyl esters by transmethylation using methanolic sodium hydroxide as described by AOAC [17]. Fatty acid methyl esters were analyzed using gas chromatography.

Cooking loss was calculated from the difference in sample mass before and after cooking (frying, grilling, microwaving and steaming).

$$\text{Cooking loss\%} = \frac{(\text{mass before cooking - mass after cooking})}{\text{mass before cooking}} \text{ x } 100$$

RESULTS AND DISCUSSION

The changes in nucleic acid nitrogen (NAN) bases content of fresh and cooked Sardine fish (mg/100g) and percentages were shown in Table (1).

The decrease percent ranged from 5% in grilled fish for 10min to 48% in boiled samplesfor 25min except of the NAN content in grilled samples for 15min increased with percentage 3%. The previous data observed that the highest content of NAN bases was 304mg/100g in grilled samples for 10min followed that the boiled for 20min compared with fresh samples was 320mg/100g whereas, the lowest content was 166mg/100g in boiled samples for 25min. The decrease percent in fried samples for 10min equal the decrease percent in Sardine samples cooked by microwave for 7min followed that the decrease percent in roasted for 25min equal in Sardine samples cooked by microwave for 5min and then, the samples grilled for 10min whereas, the NAN content increased to 3% after 15min grilling but this increment resulted in thelost of moisture and fat content during grilling on the direct fire that mean that the effect of the wet heat by boiling with rinsing and roasting more than the dry heat by grilling.

These results agree with Choi *et al.* [1] who observed that the cooking processes affect purine content, boiling high-purine foods in water can cause break-down of the purine-containing components (called nucleic acids) and eventual freeing up of the purines for absorption. Lou *et al*. [21] obtained results showed a decrease in moisture, ash and oil in grilled fish as a result of grilling process, where crude oil decreased from 22.14 to 11.40% (based on dry weight) in mullet fish. While, crude protein increased from 69.04 to 86.71% (based on dry weight) in mullet fish. Also, Lou*et al.* [3] statedthat the moist heat methods can reduce purine contents of food and the reducing effects arein the following order: boiling > steaming > microwave cooking. The changes in the purine content of tilapia (*Tilapia mossambica*) during storage, heating and drying were investigated. The content of

Table 1: Changes in Nucleic acid nitrogen bases content of fresh and cooked Sardine fish (mg/100g) and the decrease percentage (%)

Cooking duration (min)	Boiled	Fried	Grilled	Roasted	By Microwave
5	-	-	-	-	275 (14%)
7	-	-	-	-	262 (18%)
10	-	262 (18%)	304 (5%)	-	-
15	-	240 (25%)	330 (+ 3%)	-	-
20	208 (35%)	-	-	294 (8%)	-
25	166 (48%)	-	-	272 (15%)	-

Nucleic acid nitrogen bases content of fresh Sardine fish= 320 mg/100g

Table 2: Chemical composition of fresh and cooked Sardine fish (g/100g) for two cooking duration

		Boiled		Fried		Grilled		Roasted		By Microwave	
Composition analysis	Fresh	After 20min	After 25min	After 10min	After 15min	After 10min	After 15min	After 20min	After 25min	After 5min	After 7min
Moisture	68.5	65.3	63.1	66.9	65.2	63.4	61.1	65.5	62.6	61.2	57.9
Crude protein	20.2	24.7	28.7	20.7	22.6	29.8	32.1	28.3	30.0	30.3	33.8
Crude ash	1.50	1.59	1.70	1.30	1.10	1.70	2.10	1.75	1.90	1.94	2.40
Crude lipid	9.80	8.41	6.50	11.1	11.1	5.10	4.70	4.45	5.50	6.56	5.90

Table 3: Physicochemical parameters of fresh and cooked Sardine fish after the second cooking duration:

Parameters	Fresh	Boiled	Fried	Grilled	Roasted	By Microwave
T.V.N (mg/100g)	19	25	44	78	64	22
T.B.A (mg/kg)	0.92	1.67	1.96	0.89	0.77	0.66
T.M.A (mg/100g)	3.54	3.67	3.84	3.29	3.36	3.21
Cooking loss %	-	32%	23%	46%	38%	18%

these uricogenic purines in tilapia decreased 46.4 and 24.9% during 40 min boiling and steaming, respectively. However, 16.6% of purines were lost in 7 min cooking by microwave. These findings suggest that moist heat methods had a greater reducing effect on purines in tilapia than dry heat methods.

The proximate composition of fresh and cooked Sardine fish was shown in Table (2).

The highest moisture content was 68.5% in fresh samples whereas, samples cooked by microwave for 7 min recorded the lowest value 57.9% while, it contained the highest values of protein and ash reached to 33.8 and 2.4% respectively but the lowest content of them were 20.2 and 1.1% in fresh and friedSardine fish samples after 15min, respectively. Also, the fried samplescontained the highest fat content was11.1% for 10 min whereas, the lowest lipid content were 4.45 and4.70% in roasted and grilled samples after 20 and 15min, respectively.

From the previous data obtained the protein and ash content increased with increasing the cooking duration. The highest percents of protein were 30.3 and 33.8% and ash were 1.94 and 2.40% after cooking by microwave for 5 and 7min respectively compared with fresh samples were 20.2 and 1.5%. On the contrast, the moisture content decreased under the previous conditions and the fat content decreased with increasing the cooking duration were 11.1 and 10.1% after frying for 10 and 15min respectively, compared with fresh samples was 9.8%.

These results agree withMusaiger *et al*. [7] who used eight cooked species of fish and one species of shrimps (grilled, curried, fried and cooked in rice) commonly consumed in Bahrain for their proximate, mineral and heavy metal content. The results revealed that the protein content was in the range of 22.8-29.2 g/100g, while the fat content was between 2.9-11.9 g/100g. Also, different cooking treatments were used (baking, grilling, microwaving and frying), the protein and ash contents increased in all cooked fish, the fat content in fried samples was significantly increased due to absorption of fat. So, the frying method cannot be recommended for human consumption [22].

Physicochemical parameters of fresh and cooked Sardine fish after the second duration were shown in Table (3).

Total volatile nitrogen (T.V.N) of fresh and cooked Sardine fish ranged from 19 in fresh to 78 mg/100g in grilled fish whereas, both thiobarbituric acid (T.B.A) and trimethylamine (T.M.A) hadthe highest values 1.96 mg/kg and 3.84 mg/100g in fried Sardine fish respectively. Whilethe lowest values were 0.66 mg/kg and 3.21 mg/100g in samples cooked by microwave.

Both moisture and fat content of samples were lost as a result of using the different cooking methods that led to weight decrease compared with fresh samples. The cooking loss reached to 46% after grilling method for 15min followed by both roasting and boiling methods

were38 and 32% respectively and the last, the cooking loss of samples cooked by microwave was 18% compared with fresh Sardine fish.

These results agree with Dhanapal *et al.* [11] who observed that cooking of tilapia fish steaks has a significant influence on the TBA content. The levels of TBA, salt, TVBN and TPC of raw/fresh meat of tilapia, which were 0.77±0.01mg%, 0.58±0.01% and 3.57±0.01mg% changed to 1.61±0.06mg%, 0.41±0.01% and 3.59±0.01mg% respectively, after cooking.

Minerals content of fresh and cooked Sardine fish mg/100g after the second durationwereshown in Table (4).

Both grilling and cooking by microwave methods recorded the maximum values for macro-elements which reached to 269, 387 and 36 mg/100g for potassium (K), sodium (Na) and magnesium (Mg) after grilling for 15min and 164 and 346mg/100g for calcium (Ca) and phosphorous (P) after cooking by microwave for 7min whereas, the boiling method for 25min recorded the minimum values reaching to 52, 182, 264 and 22mg/100g forCa, K, Na and Mg respectively except of P was 270mg/100g after grilling for 15min compared with fresh samples were 72, 201, 308, 316 and 29mg/100g for Ca, K, Na, P and Mg, respectively.

While, the highest values for micro-elements varied with different cooking methods where they reached to 1.4 and 0.25mg/100g for iron (Fe) and cupper (Cu) after grilling for 15min, zinc (Zn) was 0.6mg/100g after frying, manganese (Mn) was 0.42mg/100g after cooking by microwave for 7min and selenium (Se) was 0.057m/100g after roasting for 25min whereas, the boiling method for 25min recorded the lowest values which were 0.7, 0.3 and 0.2mg/100g for Fe, Zn and Mn but both Cu and Se were 0.1 and 0.021mg/100g after frying for 15min.

These results agree with Musaiger *et al*. [7] who used eight cooked species of fish and one species of shrimps (grilled, curried, fried and cooked in rice) and reported that samples has a considerable content of sodium ranging from 120-600 mg/100g, potassium (310-560 mg/100g) phosphorous (200-330 mg/100g), magnesium (26-54 mg/100g) and zinc (0.4-2.0 mg/ 100g), while the other minerals arepresent to a lower extent. Also, Ersoy and Ozeren, [22] who observed that the mineral levels areaffected by cooking methods, except for Cu. Ca, K and Mg contents of grilled fish increased. However, the changes in Fe, Zn, Mn and Cu contents were found to be insignificant. Also, protein, Na, K, Ca, Mg and Zn contents of baked and microwave-cooked fish increased considerably.

Vitamins content of fresh and cooked Sardine fish after the second duration were shown in Table (5).

All vitamins soluble in fat recorded the highest values for vitamins A, D, E and K 137 IU, 327 IU, 1.7mg/100g and 2.76mcg after frying for 15min respectively, whereas, theyrecorded the lowest values 78 IU, 234 IU, 0.9 mg/100g and 1.82 mcg after boiling for 25min respectively, compared with fresh samples 108IU, 267IU, 1.3mg/100g and 2.3mcg respectively. While, the vitamins soluble in water decreased after all the cooking methods and reached to the lowest values after boiling method 32, 150, 40, 30, 4.9 and 3.8mcg compared with fresh samples 100, 300, 1130, 80, 9 and 10mcg for B1, B2, B3, B6, B12 and folic acid respectively.

These results agree with Ersoy and Ozeren, [22] who observed that the vitamin A content is significantly increased in grilled and fried fish and vitamin E is increased in all cooked fish. Vitamin B1 content of cooked fish was significantly decreased. Vitamin B2 and niacin contents of grilled fish were increased significantly. B6 content of cooked fish was significantly decreased but this did not occur for the grilled fish. High levels of vitamin and mineral contents were found in grilled catfish. However, vitamin contents were decreased, Vitamins A, E, B2, B6 and niacin were increased significantly. However, there was a decrease in vitamin B1 content. The grilling method is the most suitable method among all cooking methods.Also, Sardines are an extremely rich source of niacin. But most meats have similar amounts and it's hard to find anyone short of niacin in Canada. So we didn't want to make a fuss about it. We also have some pantothenic acid while also comprising an excellent source of vitamin A. And unless you're a vegetarian, you might not care about all our vitamin B12 [2].

Essential amino acids content of fresh and cooked Sardine fish (mg/g protein) after the second duration were shown in Table (6).

Most Essential amino acids increased after all cooking methods especially after cooking by-microwave reaching to the highest values ranged from 14 to 91 mg/g protein for tryptophan and Lucien amino acids respectively compared with both fresh samples and FAO/WHO/UNU pattern were 13 and 17 mg/g protein for tryptophan and 87 and 93 mg/g protein for Lucien respectively. Lysine and tryptophan decreased after grilling and boiling methods were 73 and 12 mg/g protein respectively compared with fresh Sardine samples and FAO/WHO/UNUpatternwere 78 and 66 mg/g protein for lysine respectively and 13 and 17 mg/g protein for tryptophan respectively. Also, the maximum value of total

Table 4: Minerals content of fresh and cooked Sardine fish (mg/100gm) after the second cooking duration:

Minerals	Fresh	Boiled	Fried	Grilled	Roasted	By Microwave
Macro-elements Calcium	72	52	61	84	111	164
Potassium	201	182	195	269	248	237
Sodium	308	264	341	387	341	375
Phosphorus	316	281	310	270	330	346
Magnesium	29	22	25	36	32	33
Micro-elements Iron	1.2	0.7	0.9	1.4	1.2	1.3
Zinc	0.7	0.3	0.6	0.4	0.3	0.5
Copper	0.25	0.21	0.10	0.25	0.19	0.18
Manganese	0.29	0.20	0.40	0.25	0.22	0.42
Selenium	0.036	0.031	0.021	0.054	0.057	0.049

Table 5: Vitamins content of fresh and cooked Sardine fish after the second cooking duration:

Vitamins	Fresh	Boiled	Fried	Grilled	Roasted	By Microwave
Soluble in fat: A (IU)	108	78	137	114	87	99
D (IU)	267	234	327	289	252	267
E (mg %)	1.30	0.90	1.70	1.59	1.29	1.52
K (mcg)	2.30	1.82	2.76	2.62	2.54	2.59
Soluble in water: B1 (mcg)	100	32	39	45	52	60
B2 (mcg)	300	150	270	210	220	280
B3 (mg)	1.13	0.04	1.03	0.73	0.05	0.09
B6 (mg)	0.10	0.03	0.08	0.06	0.05	0.07
B12 (mcg)	9	4.9	7.5	6.2	6.7	8.4
Folic acid (mcg)	10	3.8	6.9	4.1	3.9	7.1

Table 6: Essential amino acids content of fresh and cooked Sardine fish (mg/g protein) after the second cooking duration:

Essential amino acids	FAO/WHO/UNU (1989) pattern	Fresh	Boiled	Fried	Grilled	Roasted	By Microwave
Isoleucine	46	46	47	48	46	47	52
Leucine	93	87	88	88	87	89	91
Lysine	66	78	74	74	73	74	79
Methionin+ Cystine	42	32	32	31	34	33	35
Phenylalanine + Tyrosine	72	76	79	78	77	79	80
Threonine	43	44	47	44	46	46	47
Valine	55	52	52	54	53	54	56
Histedine	26	62	64	63	65	65	69
Tryptophan	17	13	12	13	12	13	14
Total E.A.A	460	490	495	493	493	500	523

essential amino acids was 523 followed 500 mg/g protein for samples cooked by microwave and roasting respectively and the minimum values were 495, 493 and 493 mg/g protein for boiling, frying and grilling compared with fresh samples and FAO/WHO/UNU pattern were 490 and 460 mg/g protein respectively [23].

Fish is known to be a source of protein rich in essential amino acids (lysine, methionine, cysteine, threonine and tryptophan). Fish muscle also contains micro- and macroelements and fat-soluble vitamins [24].

Fatty acids profile content of fresh and cooked Sardine fish (g/100g F.A) after the second duration were shown in Table (7).

Both total saturated and monounsaturated fatty acids decreased after most cooking methods were 38.12, 26.89, 33.80 and 26.68 g/100g F.A compared with fresh saturated fatty acids were44.71g/100gF.A and 29.1, 24.9, 27.6 and 21.9g/100gF.A compared with fresh monounsaturated fatty acids was 31.5g/100gF.A except of frying method increased slightly to 45.09 and 35.10g/100gF.A for saturated and monounsaturated fatty acids respectively. While total polyunsaturated fatty acids increased after all cooking methods reached to 24.99, 32.45, 28.13, 28.63 and 29.28g/100gF.A compared with fresh sardine fish was 23.58g/100gF.A that result for increasing total polyunsaturated n-6 and total polyunsaturated n-3 fatty acids where, the first increased to 9.7, 12.8, 9.6, 10.3 and 10.5g/100g compared with fresh was 9.2g/100gF.A, also, total polyunsaturated n-3 fatty acids increased to 15.0, 19.65, 18.53, 18.33 and 18.78g/100gF.A compared with fresh was 14.38g/100gF.A.

Table 7: Fatty acids profile content of fresh and cooked Sardine fish (g/100g F.A) after the second cooking duration

Fatty acids	Fresh	Boiled	Fried	Grilled	Roasted	By Microwave
Saturated fatty acids:						
C14 : 0 (Myristic)	7.1	6.4	7.8	4.1	6.90	1.98
C16 : 0 (Palmatic)	28.5	25.2	27.1	17.8	21.2	16.9
C18 : 0 (Stearic)	8.4	5.9	9.4	4.7	5.30	7.60
C20 : 0 (Arachidic)	0.71	0.62	1.79	0.29	0.40	0.20
∑ Saturated fatty acids:	44.71	38.12	45.09	26.89	33.80	26.68
Monounsaturated:						
C16 : 1 (Palmitoleic) n-7	9.3	8.5	9.8	6.7	8.9	5.6
C18 : 1 (Oleic) n-	22.2	20.6	25.3	18.2	18.7	16.3
9∑ Monounsaturated:	31.5	29.1	35.1	24.9	27.6	21.9
Polyunsaturated:						
Polyunsaturated (n-6):						
C18 : 2 (Lenoleic)	8.2	8.9	11.2	8.5	8.8	9.2
C20 : 4(Arachidonic)	1.0	0.8	1.6	1.1	1.5	1.3
∑ Polyunsaturated (n-6):	9.2	9.7	12.8	9.6	10.3	10.5
Polyunsaturated (n-3):						
C18 : 3 (Lenolenic)	1.41	1.50	1.90	2.62	2.57	2.32
C20 : 5 (EPA)	0.31	0.47	0.78	0.59	0.73	0.58
C22 : 5 (DPA)	3.77	3.82	4.90	4.52	4.13	4.58
C22 : 6 (DHA)	8.89	9.50	12.07	10.8	10.9	11.3
∑ Polyunsaturated (n-3):	14.38	15.29	19.65	18.53	18.33	18.78
∑ Polyunsaturated (n-6 + n3):	23.58	24.99	32.45	28.13	28.63	29.28
∑ Polyunsaturated / ∑Saturated	0.53	0.66	0.72	1.05	0.85	1.76
∑Monounsaturated / ∑Saturated	0.70	0.76	0.78	0.93	0.82	0.82
Ratio of ∑ n-3 / ∑ n-6	1.56	1.58	1.54	1.93	1.78	1.79
∑ EPA + DPA + DHA	12.97	13.79	17.75	15.91	15.76	16.46

EPA = Ecosapentaenoic acid
DPA = Docosapentaenoic acid
DHA = Docosahexaenoic acid

The frying method recorded the highest effect for increasing the total polyunsaturated fatty acids were 12.8 and 19.65g/100gF.A for n-6 and n-3 respectively, followed by the samples cooked by-microwave, roasting and grilling were 10.5, 10.3 and 9.6g/100gF.A for n-6 and 18.78, 18.33 and 18.53g/100gF.A for n-3 respectively, whereas, the samples boiled increased slightly to 9.7 and 15.29g/100gF.A compared with fresh Sardine fish were 9.2 and 14.38g/100gF.Afor n-6 and n-3 respectively.

The percentage between total polyunsaturated fatty acids to total saturated fatty acids ranged from 1.76 for samples cooked by-microwave to 0.66 for boiled samples compared with fresh samples was 0.53. Also, the monounsaturated to saturated fatty acids ranged from 0.93 for grilled samples to 0.76 for boiled samples compared with fresh samples was 0.70. Ratio total n-3/total n-6 ranged from 1.93 for grilled to 1.54 for fried compared with fresh samples was 1.56 whereas, the fried samples recorded the highest total EPA+DPA+DHA value was 17.75g/100gF.A compared with fresh samples was 12.97g/100gF.A which recorded the lowest value.

These results agree with Khoddami *et al*. [25] who recorded that the fatty acids in sardine wastes (Head, Intestine and Liver) arepalmitic (C16:0; 27.80- 35.56 %), stearic (C18:0; 5.90- 9.30 %), oleic (C18:1c; 15.47- 21.79 %) and docosahexaenoic acid (DHA; C22:6; 11.87- 15.95 %). The n3 / n6 ratio of the respective head, liver and intestine lipid samples showed value higher than 1. Due to n-3 fatty acid compound and n-3 / n-6 ratio, lipid from sardine waste may be a valuable source for human consumption. Cooking significantly increased MUFA content of fish fillets. The results indicated a relatively high durability of n-3 PUFAcontained in fish lipids. As a result, both conventional oven and microwave oven cooking methods can be preferred without significant loss of n-3 PUFA [8].

The study proved that some cooking methods have positive effect for decreasing NAN bases reached to 48% after boiling for 25 min where, the wet heat hashigher effect than dry heat and its effect increase with increasing the cooking duration. Also, the nutrient components increased after all cooking methodslike; Essential amino

acids especially after cooking by microwave, minerals especially after grilling, fat-soluble vitamins after frying and total polyunsaturated fatty acids increased while saturated and monounsaturated fatty acids decreased.

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