

Alteration in Fatty Acid Profiles and Formation of Some Harmful Compounds in Hammour Fish Fillets and Frying Oil Medium Throughout Intermittent Deep-Fat Frying Process

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Abstract: This research was performed to assess the alterations in fatty acid profiles for hammour fish fillets lipid and refined sunflower (RSF) oil, frying oil medium and to evaluate the formation extent of some harmful compounds including malonaldehyde, polar compounds, oxidized and trans fatty acids in the former lipids throughout intermittent deep-fat frying process. The obtained results revealed that intermittent frying process caused a significant alteration in the fatty acid profiles of hammour fish lipid and frying oil medium due to the exchange of lipid occurs throughout frying process between frying oil medium and fried fish lipid and to the oxidative degradation of unsaturated and some saturated long chain fatty acids resulting in the formation of saturated short and medium chain fatty acids. In addition, the intermittent frying conditions encouraged the formation of tested harmful compounds in hammour fish fillets and RSF oil at exceptional high levels, especially with extending the repeating use period of frying oil medium. Therefore, the present results are recommended that the repeating use period of frying oil medium throughout intermittent frying process should not exceed 16 hours to avoid the formation of harmful compounds in fried foods and frying oil medium at hazardous and toxic levels.

Key words: Harmful compounds • Fatty acid profile • Intermittent frying • Hammour Fish lipid • Frying oil medium • Fried fish

INTRODUCTION

Frying processes are considered the most commonly used procedures for the preparation, cooking and production of foods throughout the world. It is well known that fried foodstuffs, especially the fishery and meat products, are considered the most popular foods in the household and restaurants as an important item of food in the daily human diets all over the world. It is well known that during deep-fat frying process, frying fats and oils are repeatedly used at elevated temperature in the presence of the atmospheric oxygen and therefore they undergo many reactions because of heat treatment, including decomposition, oxidation, hydrolysis and polymerization [1-4]. Frying and the other thermal processes have been applied not only for technological purposes (Production of fried food with acceptable qualities) but also for safety and nutrition standpoints.

There is some evidence that highly oxidized and heated lipids may have carcinogenic properties because of potentially toxic substances which are forming as the result of the former reactions throughout frying process [2, 5-9].

Apart from this, the nutritional value of frying fats and oils is affected by the loss of polyunsaturated fatty acids, which supplement the essential fatty acids, requirement in human metabolism [6, 7, 10-12]. In this concern, many studies were carried out to try to pay more attention towards the formation of some toxic and / or carcinogenic compounds in foods as the result of cooking and the other thermal processes, these compounds are including malonaldehyde compound, the oxidized fatty acids, polar compounds and trans fatty acids as reported by Elhassaneen and Shaheen [13], Billek [14], Singh and Tayagi, [15] and Polonio *et al.* [16].

Recently, there is an increasing international concern about the toxic adversity of the former harmful compounds for human health and the formation of these compounds in foods during cooking and processing. It has been reported that the dietary intake of both the edible lipids used repeatedly for a long period as intermittent frying oil medium and fried foodstuffs containing a high level of the oxidized fatty acids, malonaldehyde, trans fatty acids and polar compounds causes a lot of pathological changes in human and mammals including the growth retardation, the reduction of feed efficiency, the impair of liver and renal functions, an enlargement of liver and kidneys and rise of the LDL-cholesterol and therefore increasing the risk in coronary heart disease [17-22]. In addition, the ingestion of frying oil medium and fried foods containing a high concentration of the former harmful compounds may be converted to be mutagenic and carcinogenic agents as the result of accumulation nature [23-26]. Thereupon, it is of great importance to perform more researches in order to throw the light on the safety of foodstuffs cooked by intermittent deep-fat frying process in Saudi Arabia. However, little efforts have been made, as well as few reports have been published in this concern.

Therefore, the present investigation is carried out to assess the alterations in fatty acid profiles and to evaluate the formation extent of some harmful and/or carcinogenic compounds including malonaldehyde, polar compounds, oxidized and trans fatty acids in hammour fish fillets lipid and refined sunflower oil, used as frying oil medium throughout the intermittent deep-fat frying process.

MATERIALS AND METHODS

Materials: Marine Hammour *Epinephelus areolatus* fishes (belong to the family Serranidae, order Perciformes and class Actinopterygii) were obtained from local market in Jeddah city, Saudi Arabia during the season of 1429-1430 AH. Whereas, the purchased fishes were washed carefully with tap water, packaged in iceboxes and transported to the Research Laboratory of Nutrition and Food Science Dept. Faculty of Education for Home Economics and Art Education, King Abd- Elaziz University, Jeddah, Saudi Arabia. Refined sunflower oil used as frying oil medium and sodium chloride were obtained from the local market in Jeddah city. All chemicals and reagents used in the analytical methods were analytical grade. Pure standards of fatty acids methyl esters used in this study were obtained from Sigma Chemical Co. P.O. box 145508, St. Louis, USA.

Methods

Preparation of Hammour Fish Fillets for Cooking by Intermittent Deep-Fat Frying Process: Fresh marine hammour fishes were dressed carefully by removing scales, fins, head, tail and viscera. The fish body was washed with tap water to remove any traces of blood and viscera. After that, the fish flesh was divided into desired consistent size fillets and then the fillets were trimmed, washed carefully by tap water, drained for a few minutes and then immersed in 10 % NaCl solution for 10 minutes. Whereas, fish fillets were washed with tap water and treated with 3 % spices' mixture (composed of black pepper powder, cumin powder and fresh garlic paste at ratio of 1:1:2, respectively). Then, the prepared fillets were used for analysis and cooking.

Intermittent Deep-Fat Frying Method: It was carried out as the most commonly cooking method applied in household and restaurants in my country. It was used for cooking the prepared hammour fish fillets according to the procedures of Tsaknis and Lalas [3] and Hend-Ganbi [19]. In this procedure, a batch of 200 g of prepared fish fillets was deep-fat fried independently in 2L of frying oil medium; refined sunflower (RSF) oil. Whereas, frying oil medium was heated in a household stainless steel pan (3 liter - Capacity, 20 cm-diameter and 15 cm-high) to a temperature of $175 \pm 5^\circ\text{C}$, then a batch of 200 g of fish fillets was fried in the heated RSF oil at previous temperature for 8-10 minutes on butane gas cooker. Frying repeat was carried out at one hour intervals for 4 hours and the total of four fryings daily were done for 6 consecutive days. At the end of fryings each day, 50 g sample of frying oil medium was removed from each fryer, filtrated, poured into tightly closed dark glass bottle - 50 ml capacity and then stored at $-18 \pm 2^\circ\text{C}$ until analyzed [20]. Also, at the end of fryings each day, an adequate portion of fried fish fillets was taken, allowed to drain until it cooled to ambient temperature and then it was ground, homogenized by electric grinder (Oster Heavy Duty Food Grinder, USA), packed in polyethylene bags and then stored at $-18 \pm 2^\circ\text{C}$ until analyzed [20]. The fryers were then cupped with their lids and the frying was continued the following day. The volume of the frying oil medium was replenished to the original volume with fresh oil at the beginning of frying in each consecutive day.

Lipid Extraction: Hammour fish fillets lipid was extracted before and after frying according to the method of Rodriguez-Estrada *et al.* [21] using a mixture of chloroform (CHCl_3) and methanol (CH_3OH) at ratio 1: 1 (v / v).

Analytical Methods: The analytical methods were included fatty acid profile analysis and determination of concentration of harmful compounds including malonaldehyde, polar compounds, the oxidized and trans fatty acids. They were carried out for fresh and fried hamour fish lipid cooked by intermittent deep-fat frying method at $170 \pm 5^\circ\text{C}$ and for fresh and heated refined sunflower oil ; used as intermittent frying oil medium , at the end of frying each day during the 6 consecutive days for 4 hours daily as the following :

Fatty Acid Profile Analysis: The fatty acid profiles of hamour fish lipid and frying oil medium samples were determined as methyl ester by gas liquid chromatography (GLC) procedure. The methyl ester samples were prepared by using BF_3 methanol as methylating agent according to the procedure of AOAC [22].

Determination of Harmful Compounds' Content: Malonaldehyde compound content (mg/kg lipid) was determined according to the procedure of Pearson [23]. Polar compounds content (%) was estimated by using chromatographic glass column method as described by Walking and Wessels [24]. The oxidized fatty acids percentage was estimated by the procedure described by AOAC [25]. Trans fatty acids content was determined (as % trieladin) according to the procedure of Bansal *et al.* [26].

Statistical Analysis: The obtained data of the harmful compounds were subjected to the analysis of variance using SAS program for the multiple comparison between the means of harmful compounds in hamour fish fillets lipid and frying oil medium formed throughout intermittent deep-fat frying process; the procedure of ANOVA and Duncan's Multiple Range Test were used according to Helwing [27]. Also, the least significant difference (L.S.D) at 0.05 level of probability was used to compare between the means of each fatty acid in hamour fish fillets lipid and frying oil medium profiles throughout the intermittent deep-fat frying process according to the procedure of Gomez and Gomez [28].

RESULTS

Fatty Acid Profiles of Hamour Fish Lipid and Frying Oil Medium (Refined Sunflower Oil) as Affected by Intermittent Deep-Fat Frying Process: As shown in Tables 1 and 2, the intermittent deep-fat frying process caused an obvious alteration in fatty acid profile of tested hamour fish fillets lipid and of refined sunflower oil; used as frying oil medium, at different rates depending upon the chemical structure and the nature of fatty acid itself and the period of tested frying process. Concerning fatty acid profile of fried hamour fillets lipid as illustrated in Table 1, it could be observed that there was a marked increase in some short chain saturated fatty acids

Table 1: Alteration in fatty acid profile (%) of hamour fish fillets lipid during intermittent deep-fat frying process

| Fatty acid | Deep-Fat Frying Period (hrs) | | | | | | | LSD at 5 % |
|------------|--------------------------------|-------|-------|-------|-------|-------|-------|------------|
| | Zero | 4 | 8 | 12 | 16 | 20 | 24 | |
| C12:0 | 0.19 | 0.27 | 0.51 | 0.86 | 1.39 | 1.74 | 2.38 | 0.15 |
| C14:0 | 5.43 | 5.61 | 6.34 | 7.29 | 7.81 | 8.49 | 9.55 | 0.78 |
| C15:0 | 3.72 | 3.95 | 4.48 | 4.83 | 5.56 | 5.92 | 6.60 | 0.43 |
| C16:0 | 12.06 | 10.18 | 11.02 | 10.57 | 11.23 | 11.78 | 12.02 | 1.29 |
| C18:0 | 9.58 | 7.93 | 6.58 | 7.61 | 8.07 | 9.65 | 10.79 | 0.67 |
| C20:0 | ND | ND | ND | ND | ND | ND | ND | 0.00 |
| C22:0 | 0.29 | 0.51 | 0.36 | 0.14 | ND | ND | ND | 0.12 |
| C14:1 | 0.43 | 0.35 | 0.21 | ND | ND | ND | ND | 0.07 |
| C16:1 | 10.08 | 8.72 | 7.57 | 6.50 | 5.19 | 4.77 | 4.06 | 0.69 |
| C18:1 | 15.16 | 17.40 | 18.23 | 17.68 | 16.26 | 15.29 | 13.71 | 1.92 |
| C18:2 | 9.40 | 14.59 | 17.46 | 20.11 | 22.08 | 19.34 | 17.50 | 2.18 |
| C18:3 | 1.89 | 1.06 | 0.68 | ND | ND | ND | ND | 0.23 |
| C20:1 | 1.35 | 1.23 | 1.05 | 0.92 | 0.79 | 0.68 | 0.53 | 0.27 |
| C20:4 | 3.51 | 3.18 | 2.90 | 2.39 | 1.96 | 1.57 | 1.33 | 0.58 |
| C20:5 | 12.16 | 10.72 | 8.29 | 7.55 | 6.37 | 7.90 | 8.85 | 1.04 |
| C22:1 | ND | ND | ND | ND | ND | ND | ND | 0.00 |
| C22:5 | 4.21 | 3.90 | 3.52 | 3.17 | 2.93 | 2.66 | 2.28 | 0.31 |
| C22:6 | 7.90 | 7.37 | 6.99 | 6.43 | 5.77 | 4.89 | 4.05 | 0.69 |
| UIFAs | 2.64 | 3.03 | 3.81 | 3.95 | 4.59 | 5.32 | 6.28 | 0.74 |
| TSFAs | 31.27 | 28.45 | 29.29 | 31.30 | 34.06 | 37.58 | 41.34 | 3.08 |
| TUFAs | 66.09 | 68.52 | 66.90 | 64.75 | 61.35 | 57.10 | 52.38 | 4.91 |
| Ks Value | 2.11 | 2.41 | 2.28 | 2.07 | 1.80 | 1.52 | 1.27 | 0.09 |

ND: Not detected, LSD at 5 %: Least significant difference (at 5% level of probability) between the levels of each fatty acid in hamour fish lipid throughout the tested frying process, UIFAs: Unidentified fatty acids, TSFAs: Total saturated fatty acids, TUFAs: Total unsaturated fatty acids, Ks value: TUFAs / TSFAs

Table 2: Alteration in fatty acid profile (%) of frying oil medium (Refined sunflower oil) during intermittent deep-fat frying process for hammour fish fillets

| Fatty acid | Deep-Fat Frying Period (hrs) | | | | | | | LSD at 5 % |
|------------|--------------------------------|-------|-------|-------|-------|-------|-------|------------|
| | Zero | 4 | 8 | 12 | 16 | 20 | 24 | |
| C12:0 | 0.12 | 0.39 | 0.60 | 0.84 | 1.28 | 1.74 | 2.38 | 0.21 |
| C14:0 | 0.36 | 0.72 | 1.05 | 1.69 | 2.42 | 3.18 | 5.05 | 0.38 |
| C15:0 | ND | 0.57 | 1.12 | 1.86 | 2.69 | 3.02 | 3.76 | 0.53 |
| C16:0 | 7.09 | 7.33 | 8.09 | 9.13 | 10.4 | 11.57 | 12.80 | 1.19 |
| C18:0 | 4.15 | 4.60 | 4.84 | 5.47 | 5.85 | 6.39 | 7.44 | 0.60 |
| C20:0 | 0.33 | 0.28 | 0.16 | ND | ND | ND | ND | 0.12 |
| C22:0 | 0.60 | 0.49 | 0.34 | 0.19 | ND | ND | ND | 0.27 |
| C14:1 | 0.29 | 0.16 | ND | ND | ND | ND | ND | 0.09 |
| C16:1 | 0.16 | 2.90 | 3.42 | 3.98 | 4.76 | 5.13 | 3.67 | 0.41 |
| C18:1 | 26.08 | 23.25 | 21.98 | 21.74 | 20.82 | 20.29 | 19.54 | 1.67 |
| C18:2 | 59.13 | 48.61 | 44.53 | 40.16 | 37.24 | 33.71 | 31.26 | 3.03 |
| C18:3 | 0.25 | 0.43 | 0.79 | 0.32 | ND | ND | ND | 0.09 |
| C20:1 | 0.17 | 0.39 | 0.62 | 0.87 | 0.93 | 1.25 | 1.58 | 0.26 |
| C20:4 | ND | 0.98 | 1.70 | 1.36 | 1.08 | 0.72 | 0.45 | 0.38 |
| C20:5 | ND | 3.02 | 3.99 | 4.50 | 5.46 | 4.87 | 3.91 | 0.74 |
| C22:1 | ND | ND | ND | ND | ND | ND | ND | 0.00 |
| C22:5 | ND | 1.74 | 2.06 | 2.45 | 2.29 | 2.03 | 1.60 | 0.57 |
| C22:6 | ND | 2.51 | 2.73 | 3.29 | 2.71 | 2.47 | 2.07 | 0.39 |
| UIFAs | 1.27 | 1.43 | 1.98 | 2.51 | 3.07 | 3.63 | 4.52 | 0.26 |
| TSFAs | 12.65 | 14.38 | 16.20 | 18.82 | 21.64 | 25.90 | 31.43 | 2.81 |
| TUFAs | 86.08 | 84.19 | 81.82 | 78.67 | 75.29 | 70.47 | 64.05 | 5.07 |
| Ks Value | 6.80 | 5.85 | 5.05 | 4.18 | 3.48 | 2.72 | 2.04 | 0.39 |

ND: Not detected, LSD at 5 %: Least significant difference (at 5% level of probability) between the levels of each fatty acid in hammour fish lipid throughout the tested frying process, UIFAs: Unidentified fatty acids, TSFAs: Total saturated fatty acids, TUFAs: Total unsaturated fatty acids, Ks value: TUFAs / TSFAs

(12:0, 14:0 and 15:0) contents and an exceptional rise in oleic acid (18:1) and linoleic acid (18:2) contents. On the contrary, the most unsaturated fatty acids content of hammour fish lipid were significantly or slightly decreased with the exception of 18:1 and 18:2 contents as mentioned before. In addition, palmitic acid (16:0) and stearic acid (18:0) exhibited a variable behavior during intermittent deep-fat frying process period. The obtained results (Table 1) also illustrated that the sum of the saturated fatty acid (SFAs) of hammour fish fillets lipid decreased from 31.27 % for fresh unfried fish lipid sample to 28.45 % for fish fillets lipid fried in frying oil medium repeatedly used for 4 hours and after that returned back to increase during the intermittent frying period. Whereas, the total SFAs percentage increased to 41.34 % at the end of frying period (after 24 hrs). On the contrary, total unsaturated fatty acids (UFAs) percentage of hammour fish fillets lipid was increased from 66.09 % for unfried sample to 68.52 % for fried fish fillets cooked in frying oil medium repeatedly used for 4 hours and after that returned back to decrease significantly into 52.38 % with intermittent frying in refined sunflower oil repeatedly used for 24 hours.

With regards the alteration in fatty acid composition of refined sunflower (RSF) oil; used as frying oil medium for cooking of hammour fish fillets as given in Table 2, it could be observed that fresh unheated RSF oil did not

contain some fatty acid such as pentadecanoic (15:0), arachidonic (20:4), eicosapentaenoic (20:5), docosapentaenoic (22:5) and docosahexaenoic (22:6) acids, while these fatty acids were found in the RSF oil repeatedly used as intermittent frying oil medium for 4 hours or more (up to 24 hours). The obtained data (Table 2) also showed that there was a marked gradual loss in some unsaturated fatty acid (14:1, 18:1 and 18:2) and long chain saturated fatty acids (20:0 and 22:0) in the RSF oil during intermittent deep-fat frying process of hammour fish fillets. Meanwhile, there was a considerable gradual increase observed in short and medium chain saturated fatty acids (12:0, 14:0, 16:0 and 18:0) and some unsaturated fatty acids (16:1 and 20:1) level. From the obtained results (Table 2), it could be also noticed that there was a significant and gradual increase in total saturated fatty acids (SFAs) with extending the repeating use period of frying oil medium throughout frying process. Whereas, the total SFAs percentage of the RSF oil was increased from 12.65 % for fresh unheated oil to 31.43 % when it was repeatedly used up to 24 hours as intermittent frying medium for cooking of hammour fish fillets. On the other hand, the sum of unsaturated fatty acids (UFAs) for the RSF oil was decreased gradually from 86.08 % for fresh unheated oil to 64.05% for the repeatedly used frying oil medium throughout intermittent frying for cooking of hammour fish fillets for 24 hours.

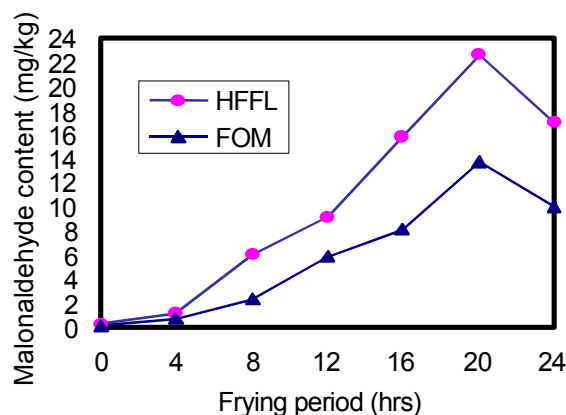


Fig. 1: Malonaldehyde content (mg/kg) in both hammour fish fillets lipid (HFFL) and refined sunflower oil as frying oil medium (FOM) as affected by intermittent deep-fat frying process

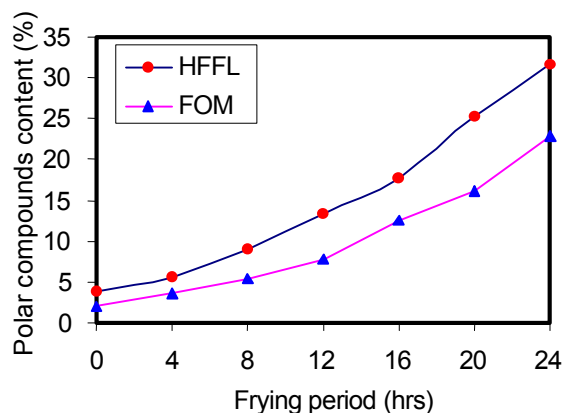


Fig. 2: Polar compounds content (%) in both hammour fish fillets lipid (HFFL) and refined sunflower oil as frying oil medium (FOM) as affected by intermittent deep-fat frying process

Formation Extent of Tested Harmful Compounds in Hammour Fish Fillets Lipid and Frying Oil Medium as Affected by Intermittent Deep-Fat Frying Process:

The formation extent of some toxic and /or carcinogenic compounds including malonaldehyd compound, polar compounds, oxidized and trans fatty acids in both hammour fish fillets lipid and refined sunflower (RSF) oil; used as frying oil medium, during the intermittent deep-fat frying process was studied and the obtained results are evident as the following.

As given in Figure 1, malonaldehyde content for fresh unfried hammour fish fillets lipid and unheated RSFOil, was 0.162 and 0.359 mg/kg lipid, respectively. The obtained results (Fig. 1) also illustrated that there was an exceptional and gradual rise in malonaldehyde content in the two tested lipids throughout the intermittent

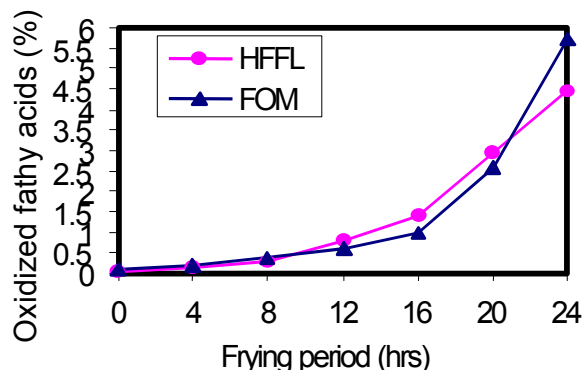


Fig. 3: The oxidized fatty acids (%) in both hammour fish fillets lipid (HFFL) and refined sunflower oil as frying oil medium (FOM) as affected by intermittent deep-fat frying process

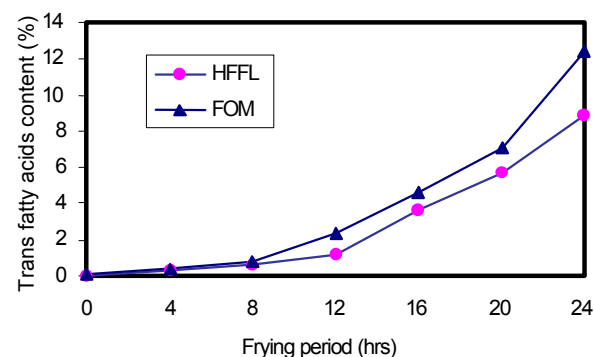


Fig. 4: Trans fatty acids (%) in both hammour fish fillets lipid (HFFL) and refined sunflower oil as frying oil medium (FOM) as affected by intermittent deep-fat frying process

deep-fat frying process with extending the period of repeating use of frying oil medium. Whereas, malonaldehyde level for the tested lipids was increased from 0.162 to 5.945 and 13.709 mg/kg for hammour fish lipid and from 0.359 to 9.237 and 22.633 mg/kg for frying oil medium (RSF oil) throughout intermittent deep-fat frying with repeating use of the RSF oil for 12 and 20 hours, respectively. After that, the former harmful compound level of the two tested lipids tended to decrease.

From the results recorded in Fig. 2, it could be observed that fresh uncooked hammour fish fillets and unheated refined sunflower oil contained 3.71 and 2.02 % polar compounds. The obtained data (Fig. 2) also illustrated that there was an exceptional gradually rise in polar compounds formation in hammour fish lipid and the RSFOil by extent of 8.5 and 11.3 folds of the corresponding fresh lipid state content after 24 hours of the repeating use of frying oil medium during intermittent

frying process. Figure 3 shows that the oxidized fatty acids (OFAs) content was increased considerably in hamour fish fillets lipid and the RSF oil with extending the repeating use period of frying oil medium (RSF oil) during intermittent frying process from 0.041 and 0.107% for fresh state to 4.47 and 3.73% for the former corresponding lipids at the end of intermittent frying period (after 24 hours). Regardless of the progressive increase in the OFAs percentage for hamour fish lipid and the RSF oil, these values were lower than the critical limit (=1%) of this criterion reported by Greene and Cumuze [29] and Saudi Standard Specifications [30] within the first 12 and 16 hours of the repeating use period of frying oil medium during intermittent frying process. After these periods, the OFAs values for tested lipids were exceeding the critical limit.

As given in Fig. 4, fresh uncooked hamour fish fillets were not contain trans isomers of unsaturated fatty acids, while fresh unheated refined sunflower (RSF) oil contained a negligible quantity (0.083%) of trans fatty acids. From the obtained results (Fig. 4), it could be also concluded that trans fatty acids in hamour fish lipid and frying oil medium (RSF oil) were formed gradually and their levels were raised exceptionally during the discontinuous frying process with extending the repeating use period of frying oil medium. Whereas, trans fatty acids percentage reached 8.83 and 12.46% in fried fish lipid and heated RSF oil at the end of intermittent frying process period (after 24 hours), respectively.

DISCUSSION

It is well known that the alteration in fatty acids composition in the edible oils and fats during thermal processing and cooking are correlated significantly with the other quality characteristics and the determination of fatty acid profile in frying oils media and fried foodstuffs lipid is providing us with the useful information about their oxidative state and health safe quality. During the intermittent deep-fat frying process, fats and oils are repeatedly used at elevated temperatures in the presence of air and moisture causing the oxidative degradation of their amino acids and the partial conversion of these lipids to volatile chain-scission products, non volatile oxidized derivatives and dimeric, polymeric or cyclic substances leading to the formation of toxic and/or carcinogenic compounds [3, 10].

As illustrated in Tables 1 and 2, a significant alterations in fatty acid profile of both hamour fish fillets lipid and refined sunflower (RSF) oil, used as frying oil

medium were taken place during intermittent deep-fat frying process; especially with extending the repeating use period of frying oil medium. These alterations in fatty acid profiles may be due to the exchange of lipid occurs throughout frying process between fried hamour fish fillets and frying oil medium [2-4, 12, 31-33] and to the oxidative degradation of unsaturated fatty acids and of some saturated long chain fatty acids resulting in the formation of saturated short and medium chain fatty acids [31, 34]. The oxidative degradation of fatty acids in the tested lipids throughout intermittent deep-fat frying process can be interpreted as follows: in saturated fatty acids, frying process may have caused an abstraction of a hydrogen atom from the active methylene group adjacent to the carboxyl group to produce free radicals followed by the oxidative degradation at α , β positions resulting in the formation of short and medium chain fatty acids, this reaction also caused a decrease in 17:0, 20:0 and 22:0 levels leading to increase of 16:0 and 18:0 contents. While, in the unsaturated fatty acids (UFAs), the double bonds migrate throughout the chain length till they occupy the α , β positions, then the oxidative degradation of unsaturated fatty acids occurs leading to decrease in 18:1, 18:2 and the other UFAs resulting in the formation of short and medium chain fatty acids, 16:0 and 18:0 levels [10, 31, 34].

It is well known that during the intermittent deep-fat frying process, frying fats and oils media are repeatedly used at elevated temperature in presence of the atmospheric oxygen and therefore they undergo many reactions, because of heat treatment, including decomposition, oxidation, hydrolysis and polymerization resulting in the formation of some potentially harmful compounds such as malonaldehyde, polar compounds, oxidized and trans fatty acids. These compounds may have mutagenic and carcinogenic properties and harmful biological effects in human and other mammals [14-16, 35].

As shown in Fig.1, the intermittent deep-fat frying process caused an exceptional and gradual rise in malonaldehyde concentration in hamour fish fillets lipid and refined sunflower (RSF) oil, used as frying oil medium, especially with extending the repeating use period of frying oil medium up to 20 hours. After that, the level of malonaldehyde in the two tested lipids was tended to decrease. The increment alteration in malonaldehyde compound concentrations in tested lipids throughout intermittent deep-fat frying process could be attributed to the secondary oxidation of fatty acids which encouraged under tested frying conditions [3, 4, 10, 31, 33, 36]. While, the reduction in this harmful compound at the end stage

of repeating use of frying oil medium (the last six hours of frying) may be due to the decomposition and volatilization of the formed secondary oxidative products [3, 33, 37]. However, malonaldehyde concentration formed in fried hammour fish fillets and frying oil medium was within the permissible limit; >10 mg/kg lipid, reported by Greene and Cumuze [29] and Saudi Standard Specifications [30] up to 16 and 12 hours of the repeating use period of frying oil medium; respectively.

With regards the formation of polar compounds throughout intermittent deep-fat frying process as illustrated in Figure 2, it could be mentioned that polar compounds were formed gradually at exceptional high extent in hammour fish fillets and frying oil medium (RSF oil) during the intermittent deep-fat frying process, especially with extending the repeating use period of frying oil medium. This observation could be attributed to the thermal effects which encourage the oxidation and polymerization of fatty acids under tested intermittent frying conditions. These finding are quite accordance with those obtained by Romero *et al.* [2], Abdulkarim *et al.* [4], Dimitra *et al.* [38] and Farhoosh *et al.* [39]. It is worth to note that polar compounds level in tested fish lipid reached the rejection level (25%) reported by Saudi Standard Specifications [30] and Romero *et al.* [40] after 16 hours of the repeating utilization of the RSF oil throughout frying process, while the level of these compounds in the RSF oil did not reach the rejection limit even at the end of frying period; after 24 hrs of the repeating use for frying oil medium.

Concerning the formation of the oxidized fatty acids during intermittent deep-fat frying conditions as exhibited in Fig. 3, it could be concluded that tested intermittent deep-fat frying conditions, including the elevated temperature and the direct contact with the atmospheric oxygen encouraged the oxidation of fatty acids in hammour fillets lipid and frying oil medium and therefore the formation of oxidized fatty acids at different rates affecting by fatty acid profile for both hammour fish lipid and frying oil medium and the period of the repeating use of frying oil medium throughout intermittent frying process. In spite of the exceptional increase in the OFAs percentages for fried fish lipid and frying oil medium, these values were within the permissible limit $\geq 1\%$, [30,41,42] until 12 and 16 hours of the repeating use of frying oil medium during intermittent deep-fat frying; respectively. The present results are in conformity with those reported by Tsaknis and Lalas [3], Abdulkarim *et al.* [4], Singh and Tayagi [15], Silva *et al.* [33] and Hassan and Abou-Arab [36].

As illustrated in Fig. 4, the formation of trans fatty acids at an exceptional high extent in hammour fish fillets lipid and refined sunflower oil, especially with extending the repeating use period of frying oil medium during the intermittent frying process may be attributed mainly to the tendency for both geometrical and positional isomerizations which are promoted by the intermittent deep-fat frying conditions [2, 26]. These results are in accordance with those obtained by Romero *et al.* [2], Stender and Dyerberg [17] and Bansal *et al.* [26]. Although it has been reported that the ingestion of high trans fatty acids level containing diets in mammals possessed carcinogenic effect and many deleterious biological functions [17, 26], the hazardous and the maximum permissible levels of these compounds until now is unknown.

Conclusion and Recomendations: From the present results, it can be concluded that the determined harmful compounds including malonaldehyde, polar compounds, the oxidized and trans fatty acids are formed at exceptional high levels in fried hammour fish fillets and refined sunflower oil, frying oil medium, throughout the intermittent deep-fat frying process especially with extending the repeating use period of frying oil medium. It is recommended that the repeating use period of frying oil medium throughout the intermittent deep-fat frying should not exceed 16 hours to avoid the deleterious effects and carcinogenic properties of the formation of tested harmful compounds at hazardous and toxic levels in fried fish and frying oil medium.

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