

# **Egyptian Journal of Chemistry**

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# Evaluation of some Functional Characteristics of Butter Oil vs. some Vegetable Oils

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#### Abstract

This study aimed at evaluating fat constants, fatty acid profile, oxidative stability, antioxidant and anti-carcinogenic activities of butter oil in comparison with some vegetable oils. The results of fat constants showed that all tested oils had good quality. The relative distribution of fatty acid profile shows that butter oil had the highest amount of butyric acid (C4:0) and lauric acid (C12:0). Palmitic acid (C16:0 saturated acid) was dominant in shortening. In contrast, sunflower and corn oils had the lowest levels of saturation. Linoleic acid (C18:2) polyunsaturated fatty acid was the most dominant in sunflower oil compared to other tested edible oils. High levels of linolenic acid (C18:3n3) fatty acid distribution was observed in soybean, cottonseed and sesame oils. The stability of butter oil and shortening under acceleration conditions of oxidation was due to their content of saturated fatty acids. Cottonseed oil had significantly the highest antioxidant and anti-carcinogenic activities compared to tested edible oils because its polyphenols contents. This study recommends using cottonseed and soybean oils in manufacturing of imitation cheese for enhancing the anti-carcinogenic activity of butter oil.

Keywords: Butter oil; soybean oil; cottonseed oil; shortening; oxidative stability; anti-carcinogenic activity

#### 1. Introduction

Dietary fats as well as oils are macronutrients and they provide human with energy and fat soluble vitamins for metabolic process [1]. However, a consumption food rich in saturated fat is directly related with hypercholesterolemia, obesity and in some cases colon cancer [2, 3]. The amount and type of fats in the diet have important effects on health of consumers especially their blood lipid profile [4].

Butter oil (ghee) is the most popular dairy product in different countries e.g. Egypt and India. It has been utilized for different purposes. These include cooking, cosmetic and medical purposes. Butter oil has many health impacts on the consumers such as source of fat soluble vitamins (A, D, E and K), essential fatty acids; conjugated linoleic and butyric acids with other lipids that have anti-colon cancer activity [5]. Furthermore, two major antioxidant compounds; vitamin E and beta carotene are found in the composition of butter oil [6]. Different types of vegetable oils replaced with milk fat in manufacturing of wide range of soft and hard chesses [7-9]. Vegetable oils are free from cholesterol with high content of unsaturated fatty acids [10]. Shea butter, olive, soybean and sunflower oils have many applications in cosmetics and they are considered as active ingredients for dermal uses [11-13]. In Egyptian markets, shortening, coconut and palm are widely used in soft cheese manufacture [9]. Shortening is solid vegetable fat, rich in Trans fatty acids and it used in both cakes and chesses [14]. Corn oil was used in manufacture of Turkish soft cheese without any negative effects on the sensory attributes [8].

However, the impact of these vegetable oils on the health of consumers still needs further investigations. Therefore, this study aims at evaluating some chemical properties, fatty acid profile, anticancer and antioxidant activities of

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Received date 10 September 2022; revised date 30 October 2022; accepted date 11 December 2022 DOI: 10.21608/ejchem.2022.161942.6959

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shortening, soybean, cotton seed, sesame, sunflower and corn oils in comparison with butter oil.

# 2. Materials and Methods

# 2.1. Materials

Un-salted butter was obtained from the Dairy Science Department, Faculty of Agriculture, Cairo University, Egypt. Shortening, Corn, Sunflower, Sesame oils were purchased from an Egyptian local market. Cotton seed oil was purchased from oil extraction plant at El Mahala El Kobra, Egypt. Soybean oil was purchased from Port Said Co. for Food Industries, Egypt. 2,2-Diphenyle-1picryhydrazyl (DPPH) was purchased from Merck, Darmstadt, Germany. MTT (3 - (4, 5 -Dimethylthiazol-2-yl)-2,5-

Diphenyltetrazolium Bromide), dimethyl sulfoxide (DMSO), trypsin and foetal bovine serum (FBS) were purchased from Sigma-Aldrich (St. Louis, USA).

#### 2.2. Methods

# 2.2.1. Preparation of butter oil

Fresh un-salted butter was heated to 60 °C then filtrated through filter paper Whatman No. 1 (Middle East Co. for Chemicals, Cairo, Egypt). The pure fat (butter oil) was reheated ( $60^{\circ}$ C) to destroy the crystal memory and was cooled to  $30^{\circ}$ C [15].

### 2.2.2. Chemical analysis

## 2.2.2.1. Determination of fat constants

Acid and peroxide values were determined according to AOAC [16]. The iodine value of samples was directly calculated from fatty acid profile using reacting ratios (calculated factors) between  $I_2$  (iodine) and either the fatty acids bound [17]. The saponification value of samples was calculated by using manual formula and either the fatty acids bound as follow:

(56.11×1000 × (% of every F.A. / 100)

M.W of every F.A

Saponification Value = -

= Total of all F.A.

#### Where, M.W = molecular weight

## 2.2.2.2. Fatty Acids Profile

Fatty acid methyl esters were prepared from total lipid by using rapid method according to the method of ISO 12966-2 [18]. Briefly, 0.1 g of the oil was placed in 5 ml screw-top test tube and 2 ml isooctane (El Nasr Co., Cairo, Egypt) was added to the tube then the tube was shaken. Methanolic potassium hydroxide solution (0.1 ml, 2N) was put on the cap fitted with a PTFE-joint, tighten the cap, and shaken vigorously for 30 seconds. The tube was left to stratify until the upper solution became clear and the upper layer containing the methyl ester was decanted. The isooctane solution is suitable for injection into the gas liquid chromatograph (GLC). Fatty acids methyl esters were injected into GLC (HP 6890 series GLC) apparatus provided with a DB-23 column (60m x 0.32mm x 25 µm). Carrier gas was N2 with flow rate 1.5 ml/min, splitting ratio of 1:50. The injector temperature was 250 °C and that of Flame Ionization Detector (FID) was 280 °C. The temperature setting was as follows 150 to 210 °C at 5°C/ min, and then held at 210°C for 25 min. Peaks were identified by comparing the retention times obtained with stander methyl esters.

#### 2.2.2.3. Oxidative stability

The stability of studied oils was determined, estimated as induction period (h) using 679 Rancimate (Metrohm, Herisau, Switzerland) at 110°C with air flow rate at 20 L/hr according to the method described by Tsaknis *et al.* [20].

#### 2.2.2.4. Antioxidant Capacity

10 mg of oils was mixed with toluenic solution of 2,2-Diphenyle-1-picryhydrazyl (DPPH) radicals and the mixture was vortexed for 20 Sec. at ambient temperature [19]. The decrease in absorption at 515 nm was measured in 1-cm quartz cells after 60 min of mixing using a UV-260 visible spectrophotometer (Shimadzu, Kyoto, Japan). The inhibition percentage was calculated from the following equation:

% Inhibition = [(absorbance of control - absorbance of test sample) / absorbance of control]  $\star$  100

## 2.2.2.5. Anti-cancer activity

The human colon cancer cell line HCT 116 was originated from the tissue of pigmented epithelium of human retina was used to study the anti-proliferative extent of tested oils on colon cancer *in vitro*. HCT 116 cell line was kindly provided by Professor Stig Linder, Oncology and Pathology department, Karolinska Institute, Stockholm, Sweden. RPMI medium was purchased from VACSERA, Giza, Egypt. This examination of anti-cancer activity depends of determination of cell viability after exposure of HCT 116 to  $10\mu g/mL$  of tested oils. The inhibition activity (%) was calculated as previously described by Hussein *et al.*[21].

## 2.2.3. Statistical analysis

A randomized complete block design with two factors was used for analysis of all data with three replications for each parameter. The treatment means were compared by Duncan test as given by Snedecor and Cochran [22].

# 3. Results and discussion

Data in Table (1) show that values of acid, peroxide and saponification values were significantly

(P < 0.05) higher in sesame oil than butter oil and other vegetable oils. Butter oil had the lowest acid and saponification values. No significant differences were found between shortening, soy bean, cottonseed, sesame, sunflower and corn oils in their saponification values. With regard to iodine value, soy bean and sunflower oils had a significant increase in the iodine values compared to corn, sesame, cottonseed, shortening and butter oils. Results obtained by Symoniuk et al. [23] showed that sunflower oil contains high degree of saturation resulting in an increase in levels of iodine value. In the same trend, Barrera-Arellano et al [24] reported that soy bean and sunflower oils had the highest unsaturation degree in comparison with different tested vegetable oils including corn oil.

Samples	Acid value (%)	Peroxide value (meq.O <sub>2</sub> /kg oil)	Saponification value (mg KOH/g oil)	Iodine value (mg KOH/g oil)
Butter oil	$0.05 \pm 0.00$ <sup>e</sup>	$1.18 \pm 0.30^{\text{ g}}$	165.83±0.52 <sup>b</sup>	$3.37{\pm}0.12^{d}$
Shortening	$0.37 \pm 0.01$ <sup>b</sup>	$1.29 \pm 0.42^{\rm f}$	207.60±0.30 <sup>a</sup>	53.12±0.41 <sup>c</sup>
Soy bean oil	$0.21\pm0.02$ <sup>c</sup>	$3.58 \pm 0.85^{\circ}$	200.23±0.61 <sup>a</sup>	138.05±0.51 <sup>a</sup>
Cottonseed oil	$0.15 \pm 0.01$ <sup>d</sup>	$8.06 \pm 1.30^{a}$	200.78±0.70 <sup>a</sup>	127.10±0.45 <sup>b</sup>
Sesame oil	$0.46 \pm 0.02$ <sup>a</sup>	$4.12{\pm}0.35^{b}$	199.54±0.65 <sup>a</sup>	125.58±0.33 <sup>b</sup>
Sunflower oil	$0.05 \pm 0.00$ <sup>e</sup>	$3.37 \pm 0.60^{d}$	199.17±0.54 <sup>a</sup>	137.50±0.27 <sup>a</sup>
Corn oil	$0.13 \pm 0.03^{d}$	$2.27 \pm 0.42^{e}$	201.25±0.51 <sup>a</sup>	125.86±0.45 <sup>b</sup>

Fatty acids profile of analyzed oils is presented in Table 2. Butter oil had the highest amount of butyric acid (C4:0) and lauric acid (C12:0). The relative distribution of fatty acid in the analysis of butter samples by Rutkowska and Adamska [25] showed the dominant of butyric acid. The maximum levels of palmitic acid (C16:0 saturated acid) were obtained from shortening and butter oils. In contrast, sunflower and corn oils had the lowest levels of saturation. This result is accordance with Symoniuk et al. [26]. High content of oleic acid (C18:1) was observed in all tested vegetable oils whereas shortening had the highest level of oleic acid. Linoleic acid (C18:2) polyunsaturated fatty acid was the most dominant in sunflower oil compared to other tested edible oils. High levels of linolenic acid

 Table 1. Fat constants of some edible oils

(C18:3n3) fatty acid distribution was observed in soybean, cottonseed and sesame oils. Cottonseed, soybean and sesame oils are characterized by high content of linolenic acid [27, 28].

Generally, the composition of fatty acids had a great impact on the oxidative stability. Oxidative stability refers to the resistance of oils/fat to oxygen and temperature and it is related to basically fatty acid composition [29]. The Rancimat method is a popular accelerated method of oxidation of tested oils/fat [30].

	Butter	Shortening	Soybean	Cotton	Sesame	Sunflower	Corn oil
Fatty acids	oil	_	oil	seed oil	oil	oil	
C4 :0	2.22	0	0	0	0	0	0
C6 :0	1.52	0	0	0	0	0	0
C8 :0	0.82	0	0	0	0	0	0
C10 :0	1.58	0	0	0	0	0	0
C12:0	2.01	0.41	0	0	0	0	0
C13:0	0	0	0	0	0	0	0
C14:0	10.10	1.16	0.07	0.28	0.06	0.08	0.33
C14 : 1	0.61	0	0	0	0	0	0
C15:0	1.34	0	0	0	0	0	0
C15:1	0.49	0	0	0	0	0	0
C16:0	30.96	44.65	11.26	14.91	10.60	7.26	12.72
C16:1	1.85	0.21	0.08	0.30	0.01	0.10	0.15
C17:0	0.87	0.09	0.11	0.10	0.08	0.042	0.09
C17:1	0.31	0.02	0.05	0.06	0.04	0.022	0.037
C18:0	12.78	4.30	4.28	3.91	5.11	3.82	2.01
C18:1	0	38.68	19.49	20.82	31.23	23.30	29.17
C18:1T	0	0	2.21	1.68	0	0	0
C18:2	0	9.69	54.07	52.34	47.96	63.91	53.91
C18:3n6	0	0.04	0.95	0.12	0.21	0	0
C18 : 3n3	0	0.16	6.55	4.51	3.67	0.28	0.70
C18:4	0	0	0	0	0	0	0
C20:0	0	0	0	0	0	0.28	0.37

**Table 2.** Fatty acids relative distribution (%) of some edible oils

Results in Figure 1 show that values of induction period (h) were significantly higher in butter oil and shortening (oils with high degree of saturation) compared to other tested edible oils. Furthermore, corn oil had the lowest oxidative stability. Generally, edible oils with the highest degree of unsaturation are more susceptible to lipid oxidation [31] whereas corn, cottonseed, soybean and sesame oils had lower induction period compared to butter and shortening oils. However, value of induction period in cream sample was 15.4 at 120 °C [32]. The stability of butter oil under acceleration conditions of oxidation was due to its content of saturated fatty acids ca. 66% [33]. Also, shortening is made by hydrogenation of vegetable oil with high degree of saturation. The most familiar saturated fatty acid profile in shortening oil is palmitic, stearic, oleic and linoleic acids [34, 35].



Figure 1. Oxidative stability of different edible oils at 110 °C expressed as induction period (h) using Rancimat

Free radical scavenging activity was measured for tested edible oils using DPPH free radical assay.

The experimental results are depicted in the Figure 2. Values of DPPH scavenging activity of corn oil and shortening were significantly much lower as compared to other tested oils. Cottonseed oil had significantly the highest antioxidant activity followed by soybean, sunflower, sesame and butter oils as well as shortening. Results obtained by Asha et al [36] showed that value of DPPH scavenging by

ghee was 35%. Antioxidant activity of cottonseed and soy bean oils was attributed to their polyphenol content [37]. Lignans, sesamin and sesamolin are responsible for the antioxidant activity of sesame oil [38]. The antioxidant activity of sunflower oil depends on method of extraction as well as type of flowers. Results obtained by Olmedo et al [39] showed that the range of DPPH scavenging (%) activity of two different sunflower oils ranged 24.6-48.2%.



Figure 2. The activity of DPPH free radical scavenging (%) of different edible oils.

Cottonseed oil had a significant anticarcinogenic activity in human colon cancer cell line HCT 116 followed by soybean and sunflower oils. In contrast, corn oil had the lowest anti-carcinogenic activity as shown in Figure 3. The anti-carcinogenic activity cottonseed and soybean oils were in good correlation with their antioxidant activity (Figure 2). Results obtained by Park et al [40] showed the anti-inflammatory and anticarcinogenic activity of cottonseed oil in mouse model. Soy bean oil is rich in isoflavones with antioxidant and anti-breast cancer activities [41]. Sunflower contains minerals such as zinc and selenium which protect from prostate cancer due to the antioxidant action [42]. Sesame oil is rich in phytases which have anti-carcinogenic activity [43]. Recently, consuming homemade ghee on an empty stomach might prevent incidence of cancer [44].



Figure 3. Anti-carcinogenic activity of different edible oils

## 4. Conclusion

Generally, fatty acid composition of different edible plays an important role in their physical-chemical properties as well as their health benefits. Cottonseed, soybean and sunflower oils had high content of omega three fatty acids with high antioxidant anticarcinogenic activities compared to other tested oils. Our results recommend blending of butter oil with cottonseed/ soybean oils for enhancing the anti-carcinogenic activity of butter oil.

# 5. Conflicts of interests

Authors declare that there are no conflicts of interests.

# 6. Acknowledgement

Authors are gratefully thanked Prof. Dr. Ahmed Aboul-Enein for his assistance in testing the anti-cancer activity of different oils.

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