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USE OF CRUDE OLIVE LEAVE JUICE AS A NATURAL ANTIOXIDANT FOR THE STABILITY OF HEATED SUNFLOWER OIL

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ABSTRACT

Olive leaves (*Kronakii* cultivar) were obtained from the annual pruning of olive trees and pressed to obtain a crude juice. An aliquots from the concentrated crude olive leave juice, represent 400, 600, 1600 and 2400 ppm as polyphenols, were added to sunflower oil. Samples of sunflower oil mixed with olive leaves juice were intermittent heated at 180 °C ± 5 for 5 hr / day and the heating process was repeated for 5 consecutive days. A control experiment was performed where butylated hydroxyl toluene (BHT) at 200 ppm was added to sunflower oil perior to intermittent heating in order to compare the antioxidant efficiency between the natural polyphenolics of olive juice and synthetic antioxidant BHT.

Some physical (refractive index, smoke point, color and viscosity) and chemical (acid value, peroxide value, iodine value, thiobarbituric acid value, saponification number, oxidized fatty acid level and polymer content) constants for the unheated and heated sunflower oil were determined. The data indicated that the addition of olive leave juice to heated sunflower oil induced remarkable antioxidant activity and at 800 ppm level were superior to that of BHT is increasing sunflower oil stability.

INTRODUCTION

Deep-fat frying is one of the most common processes used world wide for the preparation of cocked foods. It is extensively used both at home and on a commercial scale. The oils used in frying play dual functions, i.e., heating medium and enhance the formation of food flavour. Quality of oils used for frying is highly different and hence difficulties in controlling oil degradation are due to several variable such as continuous or discontinuous frying, surface-oil volume ratio, temperature, the degree of oil unsaturation and the presence of preservatives (Dobarganes and Marquez, 1996). Deep-fat frying of foods is usually performed at a high temperature (ca., 180 °C) and under atmospheric oxygen. Many studies demonstrated the occurrence of several changes in the physical and chemical characteristics of oils under frying conditions (Augustin and Berry, 1983 and Varela *et al.*, 1988).

In food a series of complex reactions occur during oil frying such as oxidation, polymerization, hydrolysis, cis / trans isomerization, double bond migration, pyrolysis and cyclization (Fritsch, 1981). Oil oxidation is one of the major deteriorative reactions occurred during frying and induce a significant loss of quality. It is well known that oil oxidation leads to changes in functional, sensory and nutritive values as well as the safety of the fried food

(Wu and Nawar, 1986). For these reasons, antioxidants are added to oil and foods rich in lipids to inhibit or depress the development of off – flavour arising from the oxidation of unsaturated fatty acids. However, the use of synthetic antioxidants such as butylated hydroxyl toluene (BHT) and butylated hydroxyl anisol (BHA) cause harmful effects on humans (Farag *et al.*, 2003a). In this connection, Chang *et al.* (1977) showed that BHT and BHA are quite volatile and easily decompose at a high temperature. In addition, these synthetic antioxidants are not effective in preventing the development of initial off-flavour

The use of natural antioxidants is highly desirable to replace the synthetic antioxidants. In this respect, the extracts of several plants have been reported to possess wide degrees of antioxidant activities (Evans and Reyhout, 1992 and Kim *et al.*, 1994). Many herbs and spices have been shown to exhibit an antioxidant activity (Farag *et al.*, 1989; Herramann, 1989; Cao *et al.*, 1996 and Jacob and Burri, 1996). The prime justification for using an antioxidant in oils is to avoid or delay oil oxidation during frying. It is worth mentioning that Farag *et al.* (2003b) extracted the polyphenolic compounds from the olive fruits and leaves and then separated into there fractions, i.e., free, esterified and bound phenolic compounds. These fractions were added separately to sunflower oil and the free phenolic compounds induced an antioxidant activity superior to that of BHT.

The present study was entailed on the direct use of juice obtained by pressing olive leaves without recourse to extraction and fractionation of the total polyphenols. Consequently, the polyphenolic compounds present in crude olive leave juice were added to sunflower oil at various levels (400, 800, 1600 and 2400 ppm) in an attempt to increase the oil stability during intermittent heating (180 °C ± 5 °C). One has to point out that the main goal of this work was to use a very cheap natural source to act as antioxidant agent. It is of interest to note that olive leaves used as a natural source for antioxidants in the present study are obtained from the annual pruning of olive plants.

MATERIALS AND METHODS

Source of olive leaves:

The ripe olive leaves of Kronakii cultivar were obtained during season 2003 from the Horticultural Research Institute, Agriculture Research center, Giza, Egypt.

Solvents and standard reagents:

All solvents used throughout the whole work were analytical grade and distilled before use. Caffeic acid (98%) was purchased from Aldrich Chemical Co. Ltd. England. Folin – Ciocalteau reagent was obtained from Gerbsaure Chemical Co. Ltd. Germany.

Sunflower oil:

Refined sunflower oil without antioxidant was obtained from Cairo and Soap Company, El-Ayat, Giza, Egypt. Peroxide and acid values for the oil were 3.98 and 0.01, respectively.

Preparation of crude olive leave juice:

Olive trees (*Kromakii* cultivar) are pruned annually at late February. The leaves of pruned branches were cleaned then pressed by hydraulic laboratory press. The resultant crude juice was concentrated using a freeze dryer and kept in a brown bottle at 5 °C until use.

General chemical analysis of olive leave juice:

Determination of moisture, ash, lipids, crude proteins (N x 6.25) and crude fibers were determined according to A.O.A.C. methods (2000). Total hydrolysable carbohydrates was determined (as glucose) after acid hydrolysis using phenol-sulfuric acid reagent (Dubois *et al.*, 1960). Total polyphenol content of crude olive leave juice was determined according to the method of Gutfinger (1981). Caffeic acid was served as a standard compound and used for the preparation of the calibration curve.

Oil heating process:

Sunflower oil (750 g) was placed in a stainless steel pan fryer (50 cm diameter x 30 cm height) and considered as a control. Portions of sunflower oil were mixed with different aliquots of crude olive leave juice to contain 400, 800, 1600 and 2400 ppm polyphenols. Also, an experiment was conducted where the oil was mixed with BHT at a concentration of 200 ppm to compare the antioxidant efficiency of the phenolics of crude olive leave juice. The various oil samples were heated continuously on a gas cooker at 180° ± 5 °C for 5 hr every day. The heating process was repeated for 5 consecutive days and stirred by hand every hour to ensure aeration and mixing. At certain periods of heating, aliquots from the oil samples were removed and stored at 5 °C for subsequent determinations.

Quality assurance tests:

The relative flow time of the various heated sunflower oil samples were measured using an Ostwald viscometer according to Joslyin (1950). Smoke point refers to temperature at which the oil sample begins to smoke and recorded as outlined by Nilson (1998). A Lovibond tintometer apparatus was applied to measure the color of the unheated and heated sunflower oil samples. The yellow glass slides were fixed at 35 and the intensity of red glasses was assigned through matching with the oil samples (Nielson, 1998). Acid, peroxide, iodine and saponification values were determined according to A.O.A.C. methods (2000). The insoluble polymers and petroleum ether insoluble oxidized fatty acids were determined according to the methods of Wu and Nawar (1986) and Billek *et al.* (1978), respectively.

RESULTS AND DISCUSSION

Egyptians are famous by eating daily several fried foods such as falafel, potatoes, egg plants, fish, chicken ... etc. Fried foods, are produced usually by heating fresh foods in an oil under atmospheric oxygen. Oxidative stability is one of the most important indicators for maintaining the quality of

edible oils. There are several ways can be used to overcome this serious oxidation problem. For instance, blending polyunsaturated vegetable oils with high oleic sunflower oil (Frankel and Huang, 1994) and the use of synthetic or natural antioxidants (Soheili *et al.*, 2002 and Ruiz *et al.*, 1999).

It has been reported that the already used synthetic antioxidants cause several deleterious effects on human health (Farag, *et al.*, 2003*a*). Hence, the natural antioxidants are the substances of choice to inhibit or suppress oil oxidation. These substances ought to be cheap and do not produce any deleterious compounds under the frying conditions. In the present study the olive leaves were pressed to obtain a juice characterized by high content of phenolic compounds. The olive leaves are obtained from the annual pruning of olive trees which is known as a waste material. Therefore, the source of natural antioxidants used in this study is priceless. Consequently, the olive leave juice is added to sunflower oil in an attempt to increase its stability during frying.

Chemical composition of crude olive leave juice:

Analysis of crude juice (moisture content 94.15 %) obtained from olive leaves of Kronakii cultivar shows that the levels of crude lipids, crude proteins, total hydrolysable carbohydrates, crude fibers and ash were: 3.71 %, 5.60 %, 70.20 %, nill and 20.50 %, respectively based on dry weight basis. These data demonstrate that the main olive juice constituent was the total hydrolysable carbohydrates, it was approximately 19 and 13 times as high as that of crude lipids and proteins, respectively. Whilst, crude proteins was about 1.5 times as great as that of crude lipids. The olive leave juice was nearly free from crude fibers. The total polyphenol content of this juice was 215 ppm determined as caffeic acid. This olive leave juice was concentrated using a freeze - dryer and used for further experiments as a natural antioxidant.

Some physical properties of heated sunflower oil mixed with various levels of phenolic compounds in olive leave juice:

A set of experiments was conducted to evaluate the antioxidant activity of the polyphenolic compounds present in olive leave juice of Kronakii cultivar. The levels of phenolic compounds added to samples of sunflower oil were 400, 800, 1600 and 2400 ppm. The control experiment refers to heated sunflower oil without the phenolic compounds of olive leave juice.

Refractive index:

Table 1 shows the changes in refractive index of heated sunflower oil at various periods. The values of refractive index of heated sunflower oil alone were gradually increased with prolonging the heating period. Mixing sunflower oil with polyphenolic compounds of olive leave juice at various levels (400 ppm, 800 ppm, 1600 ppm, and 2400 ppm.) caused gradual decrease in the refractive index values compared with values of the heated sunflower oil alone (Fig. 1). In addition, the higher level of polyphenolic compounds exhibited the highest decrease in the sunflower oil refractive index phenomenon.

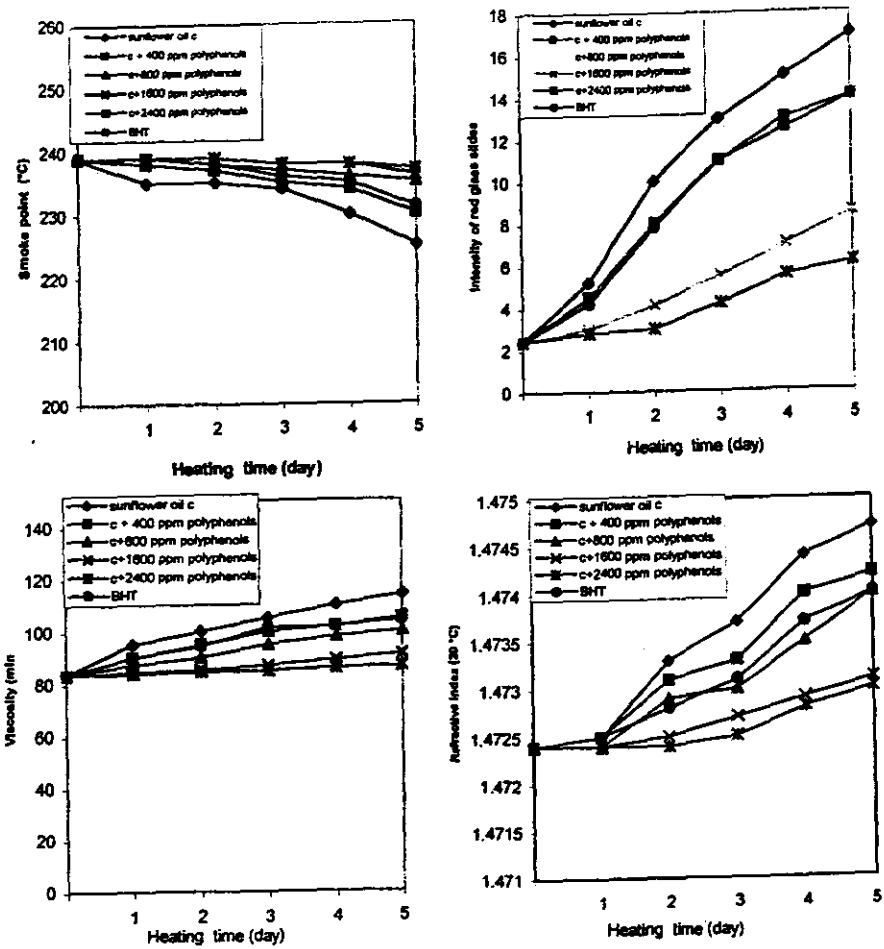

Fig. 1. Changes in some physical properties of heated sunflower oil mixed with phenolic compounds of olive leave juice and BHT.

Table (1): Changes in some physical properties of sunflower oil mixed with BHT and olive leave juice during heating (180 ± 5 °C) at various periods

Frying time (day)	Sunflower Oil (Control)	Sunflower oil BHT (200 ppm)	Sunflower oil mixed with polypenolic compounds of olive leave juice			
			400 ppm	800 ppm.	1600 ppm.	2400 ppm.
Refractive index (20 °C)						
0	1.4724^{a}	1.4724^{a}	1.4724^{a}	1.4724^{a}	1.4724^{a}	1.4724^{a}
1	1.4725^{a}	1.4725^{a}	1.4725^{a}	1.4724^{a}	1.4724^{a}	1.4724^{a}
2	1.4728^{b}	1.4728^{b}	1.4731^{b}	1.4729^{b}	1.4725^{a}	1.4724^{a}
3	1.4731^{c}	1.4781^{c}	1.4733^{bc}	1.4730^{bc}	1.4727^{b}	1.4725^{a}
4	1.4739^{d}	1.4739^{d}	1.4740^{d}	1.4735^{d}	1.4729^{bc}	1.4728^{b}
5	1.4740^{e}	1.4740^{e}	1.4742^{de}	1.4740^{e}	1.4731^{cd}	1.4730^{bc}
LSD =	0.0003	0.0001	0.0003	0.0003	0.0003	0.0003
Smoke point (°C)						
0	239^{a} + 0.62	239^{a} + 0.62	239^{a} + 0.62	239^{a} + 0.62	239^{a} + 0.62	239^{a} + 0.62
1	235^{b} + 1.09	239^{b} + 1.58	238^{b} + 2.06	239^{a} + 0.71	239^{a} + 2.31	239^{a} + 1.38
2	235^{b} + 0.18	238^{b} + 0.58	237^{c} + 0.76	238^{b} + 0.98	239^{a} + 1.03	239^{a} + 2.06
3	234^{c} + 0.23	236^{c} + 0.87	235^{d} + 0.13	237^{c} + 2.87	238^{b} + 2.06	238^{a} + 3.22
4	230^{d} + 0.42	235^{d} + 0.57	234^{e} + 0.68	236^{d} + 1.36	238^{b} + 0.17	238^{a} + 1.81
5	225^{e} + 1.31	231^{e} + 0.57	230^{f} + 0.41	235^{e} + 0.18	236^{c} + 0.89	237^{a} + 0.13
LSD =	0.87	0.71	0.87	0.87	0.87	0.87
Colour (Yellow – Red)						
0	35 - 2.4^{a}	35 2.4^{a}	35 2.4^{a}	35 2.4^{a}	35 2.4^{a}	35 2.4^{a}
1	35 5.2^{b}	35 4.3^{b}	35 4.5^{b}	35 4.0^{b}	35 3.0^{a}	35 2.8^{a}
2	35 10.0^{c}	35 7.9^{c}	35 8.0^{c}	35 7.5^{c}	35 4.1^{a}	35 3.0^{b}
3	35 13.0^{d}	35 11.1^{d}	35 11.0^{d}	35 10.0^{d}	35 5.5^{b}	35 4.2^{c}
4	35 15.0^{e}	35 12.6^{e}	35 13.0^{e}	35 12.0^{e}	35 7.0^{c}	35 5.5^{d}
5	35 17.00^{f}	35 14.1^{f}	35 14.0^{f}	35 13.5^{f}	35 8.5^{d}	35 6.1^{e}
LSD =	0.56	0.07	0.56	0.56	0.56	0.56
Viscosity (min)						
0	83.5^{a} + 0.58	83.3^{a} + 0.58	83.5^{a} + 0.58	83.5^{a} + 0.58	83.5^{a} + 0.58	83.5^{a} + 0.58
1	95.0^{b} + 1.76	90^{b} + 0.60	90.0^{b} + 1.85	87.3^{b} + 1.92	84.5^{a} + 1.07	84.0^{a} + 2.08
2	100.1^{c} + 1.05	94^{c} + 0.44	95.00^{c} + 1.76	90.0^{b} + 1.28	85.3^{a} + 1.76	84.5^{a} + 3.55
3	105.30^{d} +1.86	101^{d} +0.44	100.0^{d} + 1.76	95.0^{c} + 1.37	87.0^{b} + 1.76	85.0^{a} + 5.03
4	110.0^{e} + 2.73	102^{e} + 0.33	102.0^{e} + 1.45	98.0^{d} + 1.68	89.0^{c} + 1.45	86.0^{b} + 5.36
5	114.0^{f} + 3.05	104^{f} + 0.58	105.0^{f} + 1.76	100.0^{e} + 2.06	91.0^{d} + 1.76	
LSD =	1.83	0.62	1.83	1.83	1.83	1.83

± refers to standard error. LSD demonstrates to least significant difference test. BHT refers to butylated hydroxy toluene.

Smoke point:

The intermittent heating of sunflower oil led to a gradual and significant decrease in the smoke point values (Table 1). The addition of various levels of polyphenolic compounds of olive leave juice to sunflower oil and heated at 180 °C ± 5 °C for 5 hr / day for 5 consecutive days also caused significant decreases in smoke points (Fig., 1). In addition, the data revealed that the decrease in smoke points was related to the concentration of polyphenolic compounds. It is worth noting that the smoke points of heated sunflower oil mixed with polyphenols were generally higher than sunflower oil devoid of polyphenols.

Color:

In the present study, the values of yellow glass slides were fixed at 35 and variable values were recorded for the red glass slides. The initial red glasses value for the non-heated sunflower oil was 2.4 (Table 1). During the intermittent heating, the values of red glasses for sunflower oil were gradually and significantly increased (Fig. 1). Mixing sunflower oil with various concentrations of polyphenolic compounds of olive leave juice and heated under the aforementioned conditions, did not cause an obvious darkening in oil color.

Viscosity:

Changes in the viscosity of sunflower oil mixed with various concentrations of polyphenolic compounds of olive leave juice and heated at 180 ± 5 °C for 5 hr / day for 5 consecutive days are shown in Table 1 and depicted in Figure 1. The viscosity of sunflower oil without polyphenolic components was gradually and significantly increased during the heating process. Mixing sunflower oil with different concentrations (400 ppm, 800 ppm, 1600 ppm, and 2400 ppm) of phenolic compounds also induced gradual and significant increases in the viscosity values. The viscosity value at the end of the heating process for sunflower oil devoid of phenolic compounds was about 1.08, 1.14, 1.25 and 1.32 times are great as that of sunflower oil mixed with 400, 800, 1600 and 2400 ppm, of polyphenolic compounds, respectively. This means that the highest level of polyphenols added to sunflower oil induced the least change on sunflower oil viscosity.

Acid value:

The data in Table 2 and illustrated in Fig. 2 indicated that acid value of sunflower oil increased significantly during heating and was strongly correlated with prolonging the heating period. The addition of various levels of polyphenolic compounds of olive leave juice caused a depression in the acid value during the heating process. The acid value of heated sunflower oil at the end of heating process was about 0.91, 1.04, 0.41, and 0.14 times as great as that of sunflower oil mixed with 400, 800, 1600 and 2400 ppm polyphenols, respectively. These results indicate that polyphenolic compounds decreased the oil hydrolytic rancidity. In this respect, Farag *et al.* (2003b) reported that total and free polyphenols obtained from both leaves and fruits of Kronakii olive cultivar possessed antihyrolytic activity and increased with concentration.

Peroxide value:

The results demonstrate the occurrence of gradual increases in the peroxide value of the heated sunflower oil alone and mixed with all levels of polyphenolic compounds (Table 2 and Fig. 2). In addition, the higher level of polyphenolics induced the highest antioxidant activity. In fact, the peroxide value of heated sunflower oil mixed with 2400 ppm polyphenolics at the end of heating process was about 11.3, 9.2, 2.8 and 11.4 times as low as that for sunflower oil mixed with 400, 800. 1600 ppm and control, respectively.

The acceleration of sunflower oil oxidation due to heating at 180 ± 5°C may be interpreted as follows. The first step of lipid oxidation is the abstraction of a hydrogen atom from the active methylene group to form free radical (Farag *et al.*, 2003b). This reaction can be accelerated by the addition of a radical source, by light or by raising the temperature. As expected heating at 180° ± 5 °C for 5 hr / day and for 5 consecutive days is sufficient to produce free radicals which in turn rapidly react with the atmospheric oxygen to produce hydroperoxids. It appears that there is a relationship between the

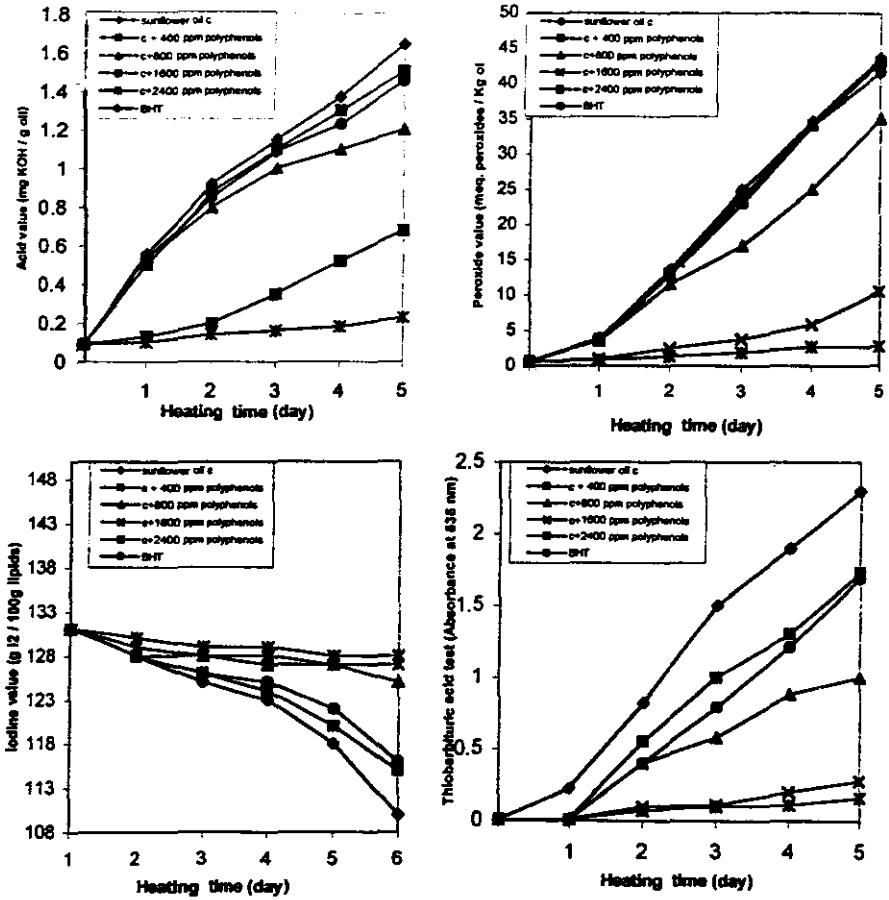

Fig. 2. Changes in acid, peroxide, thiobarbituric acid (TBA) and iodine values of heated sunflower oil mixed with phenolic compounds of olive leave juice and BHT.

Table (2): Changes in some chemical properties of sunflower oil mixed with BHT and olive leave juice during heating (180 ± 5 °C) at various periods

Frying time (day)	Sunflower oil (control)	Sunflower oil BHT (200 ppm)	Sunflower oil mixed with polyphenolic compounds of olive leave juice			
			400 ppm.	800 ppm.	1600 ppm.	2400 ppm.
Acid value (mg KOH/g oil)						
0	$0.09^{a} \pm 0.01$	$0.09^{a} \pm 0.01$	$0.09^{a} \pm 0.01$	$0.09^{a} \pm 0.01$	$0.09^{a} \pm 0.01$	$0.09^{a} \pm 0.01$
1	$0.56^{b} \pm 1.76$	$0.54^{b} \pm 0.04$	$0.50^{b} \pm 0.02$	$0.45^{b} \pm 0.02$	$0.13^{b} \pm 0.04$	$0.10^{a} \pm 0.06$
2	$0.92^{c} \pm 0.01$	$0.85^{c} \pm 0.03$	$0.88^{c} \pm 0.01$	$0.80^{c} \pm 0.02$	$0.20^{c} \pm 0.01$	$0.14^{b} \pm 0.02$
3	$1.15^{d} \pm 0.30$	$1.09^{d} \pm 0.04$	$1.10^{d} \pm 0.03$	$1.00^{d} \pm 0.10$	$0.35^{d} \pm 0.01$	$0.16^{bc} \pm 0.02$
4	$1.37^{e} \pm 0.34$	$1.23^{e} \pm 0.05$	$1.3^{e} \pm 0.03$	$1.10^{e} \pm 0.16$	$0.52^{e} \pm 0.02$	$0.18^{cd} \pm 0.010$
5	$1.64^{f} \pm 0.33$	$1.45^{f} \pm 0.4$	$1.50^{f} \pm 0.03$	$1.20^{f} \pm 0.206$	$0.68^{f} \pm 0.02$	$0.23^{e} \pm 0.05$
LSD=	0.03	0.05	0.03	0.03	0.03	0.03
Peroxide value (meq.peroxides/Kg oil)						
0	$0.60^{a} \pm 0.06$	$0.60^{a} \pm 0.06$	$0.60^{a} \pm 0.06$	$0.60^{a} \pm 0.06$	$0.60^{a} \pm 0.06$	$0.60^{a} \pm 0.06$
1	$3.90^{b} \pm 0.52$	$3.90^{b} \pm 0.55$	$3.67^{b} \pm 0.29$	$3.60^{b} \pm 0.88$	$0.98^{a} \pm 0.30$	$0.78^{a} \pm 0.72$
2	$13.61^{c} \pm 0.63$	$12.9^{c} \pm 0.49$	$13.00^{c} \pm 0.58$	$11.50^{c} \pm 0.68$	$2.50^{a} \pm 0.58$	$1.30^{a} \pm 0.19$
3	$17.21^{d} \pm 0.28$	$19.10^{d} \pm 1.28$	$17.00^{d} \pm 0.73$	$17.00^{d} \pm 0.55$	$3.81^{ab} \pm 0.73$	$1.92^{a} \pm 0.44$
4	$25.00^{e} \pm 0.67$	$34.00^{e} \pm 0.35$	$24.00^{e} \pm 0.79$	$25.00^{e} \pm 0.35$	$5.85^{bc} \pm 0.30$	$2.71^{b} \pm 0.59$
5	$43.50^{f} \pm 0.37$	$41.50^{f} \pm 0.47$	$43.00^{f} \pm 0.79$	$35.00^{f} \pm 0.32$	$10.50^{d} \pm 0.27$	$3.80^{bc} \pm 0.25$
LSD =	0.06	0.8	0.06	0.06	0.06	0.06
Iodine value (g I_2 / 100g lipids)						
0	$131.00^{a} \pm 2.21$	$131.00^{a} \pm 2.21$	$131.00^{a} \pm 2.21$	$131.00^{a} \pm 2.21$	$131.00^{a} \pm 2.21$	$131.00^{a} \pm 2.21$
1	$128.00^{b} \pm 3.06$	$128.00^{b} \pm 0.58$	$128.00^{b} \pm 3.05$	$128.00^{b} \pm 2.91$	$129.00^{b} \pm 2.68$	$130.00^{a} \pm 3.60$
2	$125.00^{c} \pm 2.98$	$126.00^{c} \pm 0.33$	$126.00^{c} \pm 2.69$	$128.00^{b} \pm 2.82$	$128.00^{c} \pm 3.05$	$129.00^{ab} \pm 3.14$
3	$123.00^{d} \pm 3.22$	$125.00^{d} \pm 0.58$	$124.00^{d} \pm 2.67$	$127.00^{bc} \pm 2.01$	$128.00^{c} \pm 3.05$	$129.00^{ab} \pm 2.72$
4	$118.00^{e} \pm 0.73$	$122.00^{e} \pm 0.58$	$120.00^{e} \pm 3.17$	$127.00^{bc} \pm 2.94$	$127.00^{d} \pm 2.52$	$128.00^{c} \pm 3.29$
5	$110.00^{f} \pm 5.29$	$116.00^{f} \pm 0.58$	$115.00^{f} \pm 2.07$	$125.00^{d} \pm 2.09$	$127.00^{d} \pm 3.06$	$128.00^{c} \pm 3.72$
LSD=	1.06	0.64	1.06	1.06	1.06	1.06
Thiobarbituric acid test (Absorbance at 535 nm)						
0	$0.01^{a} \pm 0.16$	$0.01^{a} \pm 0.16$	$0.01^{a} \pm 0.16$	$0.01^{a} \pm 0.16$	$0.01^{a} \pm 0.16$	$0.01^{a} \pm 0.16$
1	$0.23^{b} \pm 0.11$	$0.013^{b} \pm 05$	$0.02^{a} \pm 0.18$	$0.01^{a} \pm 0.17$	$0.01^{a} \pm 0.15$	$0.01^{a} \pm 0.13$
2	$0.82^{c} \pm 0.32$	$0.6^{c} \pm 0.05$	$0.55^{b} \pm 0.18$	$0.40^{b} \pm 0.19$	$0.01^{a} \pm 0.10$	$0.07^{a} \pm 0.13$
3	$1.50^{d} \pm 0.28$	$1.79^{d} \pm 0.01$	$0.80^{c} \pm 0.16$	$0.58^{c} \pm 0.10$	$0.11^{b} \pm 0.14$	$0.10^{ab} \pm 0.18$
4	$1.9^{e} \pm 0.31$	$1.21^{e} \pm 0.01$	$1.30^{d} \pm 0.19$	$0.88^{d} \pm 0.13$	$0.19^{bc} \pm 0.17$	$0.11^{bc} \pm 0.16$
5	$2.30^{f} \pm 0.54$	$1.69^{f} \pm 0.06$	$1.73^{e} \pm 0.17$	$1.00^{e} \pm 0.17$	$0.28^{d} \pm 0.18$	$0.16^{cd} \pm 0.36$
LSD=	0.07	0.04	0.07	0.07	0.07	0.07

± refers to standard error. LSD demonstrates to least significant difference test.

BHT refers to butylated hydroxy toluene.

antioxidant efficiency and the chemical composition of phenolic compounds. The main structural feature required for antioxidant activity is a phenolic ring containing hydroxyl groups. The evidence for this structural requirement is supported by the powerful anti-oxidant activities of the well-known synthetic BHT and the natural antioxidant thymol (Farag *et al.*, 1989 and Topallar *et al.*, 1997). One would relate the anti-oxidant activity of BHT or thymol and other phenolic substance to the inhibition of hydroperoxide formation through donation of hydrogen atom from the OH group of the phenolic compounds to the lipid radical which in turn produce a stable product.

Several authors isolated phenolic compounds from different sources and exhibited an antioxidant activity. In this respect, Toda *et al.*, (1996) and Chuda *et al.*, (1996) recorded an antioxidative phenolics isolated from food plants such perilla and garland, respectively. Plant flavonoids have been shown to be powerful antioxidants (Vinson *et al.*, 1998). The data of the aforementioned authors support the findings of the present study.

Thiobarbituric acid value (TBA):

The results of TBA test indicate the incidence of gradual and significant increases on the TBA values for the heated sunflower oil. Also, mixing sunflower oil with various concentrations of olive leave juice polyphenols exhibited a reduction in the secondary oxidation products (Table 2 and Fig. 2). The data for TBA values at the end of heating process were approximately 1.3, 2.3, 8.2 and 14 times as low as that for heated sunflower oil mixed with 400, 800, 1600 and 2400 ppm, respectively. This means that the polyphenolic compounds of olive leave juice at the highest level produced the lowest level of TBA reacting substance.

Comparing the antioxidant efficiency of BHT at 200 ppm with the various levels of phenolic compounds of olive leave juice, one would observe that the natural phenolic compounds at 400 ppm possessed nearly the same antioxidant activity as that of BHT at 200 ppm. whilst, 800 ppm level exhibited antioxidant activity superior to that of BHT in retading sunflower oil oxidative rancidity (Tables 1 and 2). Therefore, one would recommend to add 800 ppm of the phenolic compounds of olive leave juice to increase oil stability.

Iodine value:

The data in Table 2 and Figure 2 demonstrate that iodine values of heated sunflower oil were gradually decreased by prolonging the heating process. However, mixing sunflower oil with 1600 ppm and 2400 ppm polyphenolic compounds of olive leave juice induced the least decrease in iodine value. One would expect to achieve these results, since the phenolic compounds remarkably lower the rate of early stages of sunflower oil oxidation.

Oxidized fatty acids:

The initial value of oxidized fatty acids in fresh sunflower oil was 0.1 %. Upon heating sunflower oil under the aforementioned conditions, the levels of oxidized fatty acids were gradually and significantly increased throughout the heating period (Table 2 and Figure 3). The levels of oxidized fatty acids of sunflower oil mixed by phenolic compounds were also increased during the heating process. However, the levels of oxidized fatty acids in this case were lower than that of the oxidized fatty acid content of heated sunflower oil alone. In fact, heated sunflower oil alone had the higher level of oxidized fatty acids being about 1.1, 1.3, 3.3, 3.6 and 12.7 times as great as that of sunflower oil catalyzed by 400, 800, 1600 and 2400 ppm at the end of heating period, respectively.

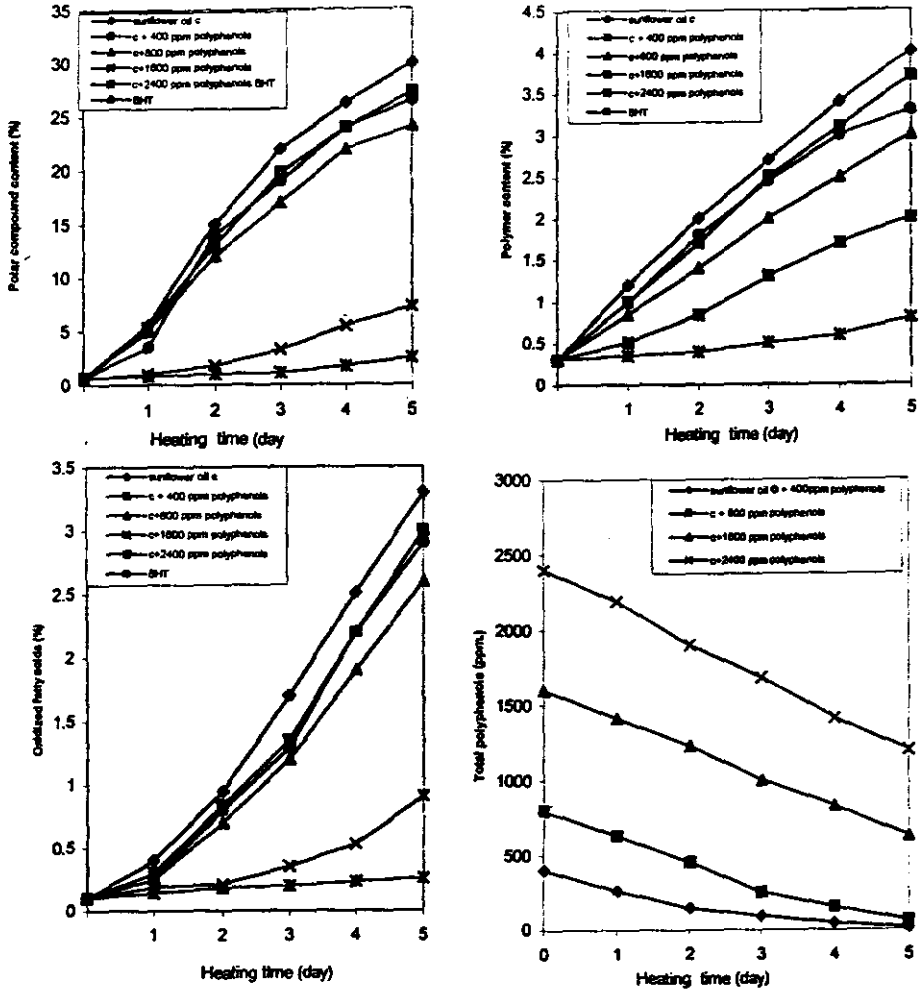

Fig.3. Changes in polar compound, polymer compound, oxidized fatty acid and total phenolic compound contents of heated sunflower oil mixed with phenolic compounds of olive leave juice and BHt.

Polar compounds:

Table 3 indicates that the levels of polar compounds of heated sunflower oil were gradually and significantly increased towards the end of heating process. The level of polar compounds at the end of the experiment was approximately 49 times as great as that at the beginning of the heating experiment (Table 3). The value of polar compounds at the end of heating period of sunflower oil mixed by 400, 800, 160 and 2400 ppm polyphenolic substances were about 0.91 0.80, 0.24 and 0.08 times as that heated sunflower oil alone, respectively. This finding indicate the powerful antioxidant activity of the olive juice polyphenols and was entirely dependent upon their concentration.

Polymer content:

The initial polymer content of fresh sunflower oil was 0.30 % and this value was increased progressively with heating period (Table 3). The changes in polymer contents of heated sunflower oil mixed by various concentrations of polyphenolics of olives leave showed linear increases with time (Figure 3). The slope values were 0.34, 0.27, 0.16 and 0 04 for heated sunflower oil mixed with 400, 800, 1600 and 2400 polyphenolic compounds of olive leave juice, respectively. This means that as the level of polyphenolic increased the formation of polymers decreased.

Polyphenolic compounds:

Changes occurring in the concentrations of polyphenolic compounds of olive leave juice added to sunflower oil and heated intermittently (180 °C ± 5 °C , 5 hr / day , for 5 consecutive days) are shown in Figure 3. The levels of phenolic compounds were linearly and significantly decreased during the entire heating process. The lost amounts of the polyphenolic compounds were 388, 740 , 975 and 1200 ppm at the end of the heating process when sunflower oil samples mixed by 400, 800. 1600 and 2400 ppm, respectively. These findings indicate that there was an inverse correlation between the amounts of polyphenolic lost during heating and the stability of sunflower oil.

In this respect, the changes in tocopherol content of oil used for deep fat frying of potatoes were studied by Gordon and Kourimska (1995). It has been found that *a*- tocopherol was lost much faster than â, م or ن-tocopherol, with a reduction of 50 % *a* -tocopherol after 4-5 frying operations compared with values of a bout 7, and 7- 8 frying operations for â and م tocopherol, respectively. Also, Ruiz *et at.* (1999) found that the level of *a* - tocopherol was lost during frying. These results are in line with data of the present study.

As already mentioned the olive leave juice contained polyphenolic compounds (215 ppm.). The antioxidant effect of this juice on sunflower oil stability was related to the presence of the polyphenolic compounds and this phenomenon was entirely depend upon their concentration. One would like to characterize the phenolic components of olive leave juice. However, the lack of the authentic substances prevented the identification of the olive leave juice components. In this respect, Benavente-Gareia *et al.* (2000) indicated the presence of oleuropein, followed by hydroxy tyrosol, the flavone -7-

glucosides of luteolin and apigenin, and verbascoside (a conjugated glucoside of hydroxyl tyrosol and caffeic acid) in olive leaf extract. The outcome of the present study suggests that crude olive juice can be used in practical operation to extend the frying life of frying oils.

Table (3): Changes in some chemical properties of sunflower oil mixed with BHT and olive leave juice during heating (180 ± 5 °C) at various periods

Frying time (day)	Sunflower oil (control)	Sunflower oil BHT (200 ppm)	Sunflower oil mixed with polyphenolic compounds of olive leave juice			
			400 ppm.	800 ppm.	1600ppm.	2400 ppm.
Polar compound content (%)						
0	0.61^{e} + 0.29	0.61^{e} + 0.29	0.61^{e} + 0.29	0.61^{e} + 0.29	0.61^{e} + 0.29	0.61^{e} + 0.29
1	5.61^{c} + 0.30	5.30^{c} + 0.01	5.40^{c} + 0.32	5.00^{c} + 0.51	1.00^{e} + 0.31	0.81^{e} + 0.51
2	15.00^{b} + 0.58	15.5^{b} + 0.44	13.00^{b} + 0.73	12.00^{b} + 0.52	1.78^{e} + 0.72	1.00^{e} + 0.59
3	20.60^{ab} + 0.40	19^{ab} + 0.17	19.75^{ab} + 0.92	17.00^{ab} + 0.33	3.30^{de} + 0.96	1.13^{e} + 0.39
4	26.30^{a} + 0.67	24^{a} + 0.00	24.00^{a} + 0.79	22.00^{a} + 0.60	5.51^{d} + 0.79	1.75^{e} + 0.12
5	30.00^{a} + 0.95	26^{a} + 0.26	27.30^{a} + 1.10	24.10^{a} + 0.57	7.30^{bc} + 1.10	2.50^{de} + 0.17
LSD=	2.31	0.27	2.31	2.31	2.31	2.31
Polymer content (%)						
0	0.30^{a} + 0.11	0.30^{a} + 0.11	0.3^{a} + 0.11	0.30^{a} + 0.11	0.30^{a} + 0.11	0.30^{a} + 0.11
1	1.20^{a} + 0.13	1^{a} + 0.6	1.0^{a} + 0.13	0.85^{a} + 0.20	0.51^{a} + 0.13	0.35^{a} + 0.24
2	2.01^{a} + 0.22	1.8^{a} + 0.6	1.70^{a} + 0.16	1.40^{a} + 0.36	0.84^{a} + 0.16	0.39^{a} + 0.48
3	2.70^{a} + 0.19	2.45^{a} + 0.6	2.50^{a} +0.16	2.00^{a} + 0.29	1.30^{a} +0.22	0.50^{a} + 0.44
4	3.40^{a} + 0.22	3^{a} + 0.12	3.10^{a} +0.26	2.50^{a} + 0.26	1.10^{a} + 0.26	0.59^{a} + 0.22
5	4.00^{a} + 0.20	3.30^{a} + 0.43	3.70^{a} + 0.31	2.60^{a} + 0.53	2.0^{a} + 0.30	0.80^{a} + 0.80
LSD=	0.09	0.23	0.09	0.09	0.09	0.09
Oxidized fatty acids (%)						
0	0.10^{e} + 0.05	0.10^{e} + 0.05	0.10^{e} + 0.05	0.10^{e} + 0.05	0.10^{e} + 0.05	0.10^{e} + 0.05
1	0.40^{e} + 0.03	0.25^{e} + 0.02	0.30^{e} + 0.01	0.25^{e} + 0.01	0.19^{e} + 0.04	0.14^{e} + 0.01
2	0.95^{c} + 0.06	0.86^{c} + 0.06	0.84^{c} + 0.04	0.70^{c} + 0.02	0.21^{de} + 0.03	0.18^{de} + 0.02
3	1.70^{b} + 0.02	1.29^{b} + 0.12	1.20^{b} + 0.04	1.20^{b} + 0.04	0.35^{d} + 0.01	0.20^{de} + 0.02
4	2.51^{a} + 0.02	2.26^{a} + 0.15	2.20^{a} + 0.02	1.90^{a} + 0.03	0.53^{c} + 0.01	0.23^{de} + 0.05
5	3.30^{a} + 0.03	3.90^{a} + 0.13	3.00^{a} + 0.03	2.60^{a} + 0.06	0.91^{c} + 0.02	0.26^{de} + 0.05
LSD=	0.05	0.12	0.05	0.05	0.05	0.05

± refers to standard error. LSD demonstrates to least significant difference test.
BHT refers to butylated hydroxy toluene.

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استخدام عصير أوراق الزيتون الخام كمادة طبيعية مضادة للأكسدة لزيادة ثبات زيت عباد الشمس المسخن

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جمعت أوراق الزيتون صنف كروناكى يدويا من أشجار الزيتون وتم كبسها للحصول على عصير الأوراق الخام. تم تركيز العصير الخام بالتجفيد. وحضرت تركيزات من العصير المحتوى على مركبات عديدة الفينول (٤٠٠– ٦٠٠ – ١٦٠٠ – ٢٤٠٠ جزء في المليون) وأضيفت إلى زيت عباد الشمس. سخن الزيت المضاف له التركيزات السابقة على حرارة ١٨٠° م ± ٥° م لمدة ٥ ساعات يوميا واستمرت المعاملة ٥ أيام متتالية.

وفى تجربة أخرى أضيفت مادة البيوتاليتد هيدروكسى تولوين المضادة للأكسدة المخلقة إلى زيت عباد الشمس بتركيز ٢٠٠ جزء في المليون وسخن الزيت أيضا بنفس المعاملة السابق ذكرها. تم تقدير عدد من الخواص الطبيعية لزيت عباد الشمس المسخن وغير المسخن مثل معامل الانكسار – نقطة التدخين – اللون– الكثافة. كذلك قدرت مجموعة من الثوابت الكيميائية مثل رقم الحموضه رقم البيروكسيد رقم حمض الثيوباربيتيوريك. كذلك المحتوى من الأحماض المؤكسدة والمواد المتبلمرة. وأوضحت النتائج أن إضافة عصير أوراق الزيتون إلى زيت عباد الشمس المسخن إن له خواص مضادة للأكسدة وتبين إن تركيز ٨٠٠ جزء في المليون هو التركيز الموصى اضافتة للزيت. وأوضحت أيضا الدراسة باستبعاد استخدام مادة بيوتاليتيد هيدروكسى تولوين كمادة مضادة للأكسدة نظرا لما لها من تأثيرات ضارة على صحة الإنسان .