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Assesment of Total Polyphenolic Contents in Virgin Olive Oil Consumed in Egypt

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ABSTRACT **:** ***This study assesses the level of total polyphenol in certain samples of Egyptian Virgin and Extra virgin olive oil by a UV spectrophotometer. A diluted olive oil sample or phenolic standard was blended with acqueous Na2CO3 and Folin-Ciocalteau reagent and the concentrations of total polyphenols were evaluated spectormetrically at 765 nm. the solutions of Gallic acid criterion were utilized to calibrate the system. it was found the concentrations of total polyphenols in the measured olive oil samples varies from 0 to286 mg/kg. It was found that most of concentrations of the analyzed virgin and extra virgin olive oil samples fall within higher total phenolic content. It was found that few numbers of analyzed olive oil samples contain low concentrations of polyphenol content. Since the the beneficial health aspects of virgin or extra virgin olive oil are strongly related to phenol content we recommend there should be quality control on samples with low polyphenol content that are labeled and sold as virgin or extra virgin olive oil.***

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1.Introduction

Virgin olive oil is distinctive amongst all vegetable oils due to its excessive content of phenolic compounds. due to the fact phenols are the most essential antioxidants that make contributions substantially to the stableness of the oil and inhibit many illnesses, their composition could be used to assess the quality of the oil (Tura et al., 2007). Virgin olive oil is extracted mechanically from olive fruits. Furthermore, phenol compounds have excellent antioxidant, against-cancer, and against-inflammatory properties, making them suitable in preventing in these diseases. Moreover, the effect of phenolic compounds on the sensory properties of virgin olive oil should be highlighted. Specifically, the compounds of polyphenol are related to the taste of bitterness and tangy flavor in oil(Bendini et al., 2007). In olive oil, phenolic compounds, also known as polyphenols in the literature, are a unique combination of various chemical compounds. Many factors influence phenolic compound concentration and composition in virgin olive oil, the most important of which are the variety, agricultural area, level of maturation of the fruit and the way of manufacturing the oil (Lerma-García et al., 2009). As per literature, the phenolic compound content of olive oil varies from 40 to 1000 mg/kg (Serville et al., 2002). Carotenes and phenolic components, which include lipophilic and hydrophilic phenols, are the primary antioxidants in virgin olive oil. some hydrophilic in virgin olive oil are not found in other oils and fats, whereas lipophilic phenols, including tocopherols, can present in other vegetable oils(Houshia et al.,2014). various agriculture and technological factors influence the phenol content of virgin olive oil. Because of the presence of a catechol group of phenolic content such as hydroxytyrosol and derivatives secoiridoids, this oil has a significantly longer shelf life than other vegetable oils(Šarolić et al.,2014). Oleuropein, a heterosidic ester of elenolic acid with 3,4-dihydroxyphenethylalcohol (hydroxytyrosol), is the main phenolic compound in olive fruit. Other glycosides present in the fruit involve ligstroside, an elenolic acid heterosidic ester with 4-hydroxyphenethyalcohol (tyrosol), and verbascoside, a caffeic acid heterosidic ester with hydroxytyrosol flavonols and Flavones (luteolin-7-glycoside, apigenin-7-glycoside, luteolin-5-glycoside) (Houshia et al.,2014). Because virgin olive oil contains a high concentration of antioxidant polyphenols (hydroxytyrosol and

oleuropein), it is comparatively steady and relcutant to oxidation, lowering the creation of harmful carcinogenic lipid peroxidation products during storage. Furthermore, polyphenols found in olive oil are known to promote positive genetic changes. Numerous studies in human cancer cell lines have confirmed oleuropein's anticancer activity, including breast adenocarcinoma (cell lines MCF-7, MDA), melanoma (cell line RPMI 7951), urinary bladder carcinoma (cell line T-24), colorectal adenocarcinoma (cell lines HT 29, Caco-2, LoVo), prostate cancer and (cell line 786-O) renal cell adenocarcinoma (Gorzynik-Debicka et al.,2018). Phenolic compounds found in VOO have been shown to protect against LDL oxidation. The European Food Safety Authority (EFSA) published a statement in November 2011 about the advantages of consuming phenolic-rich olive oil on a daily basis, like virgin olive oil (Planell et al., 2014). 2 tablespoons of Extra virgin olive oil (EVOO) per day could lower the risk of coronary diseases by promotnig blood lipid and lipoprotein levels. Higher phenol content EVOO (at least 150 mg/kg) could decrease LDL (bad cholesterol), whereas the lower total phenol content (refined olive oil) may increase LDL (bad cholesterol) (Flynn et al., 2015). Because of various technological operations, the total phenolic content of olive oil may vary. Figure 1 depicts the concentrations of the total polyphenol in olive oil as a function of the olive oil production process. Because some precision components of olive oil are solvable in water , the content of phenolic compounds found in olive oil is greatly influenced by the extraction process (Gorzynik-Debicka et al.,2018).

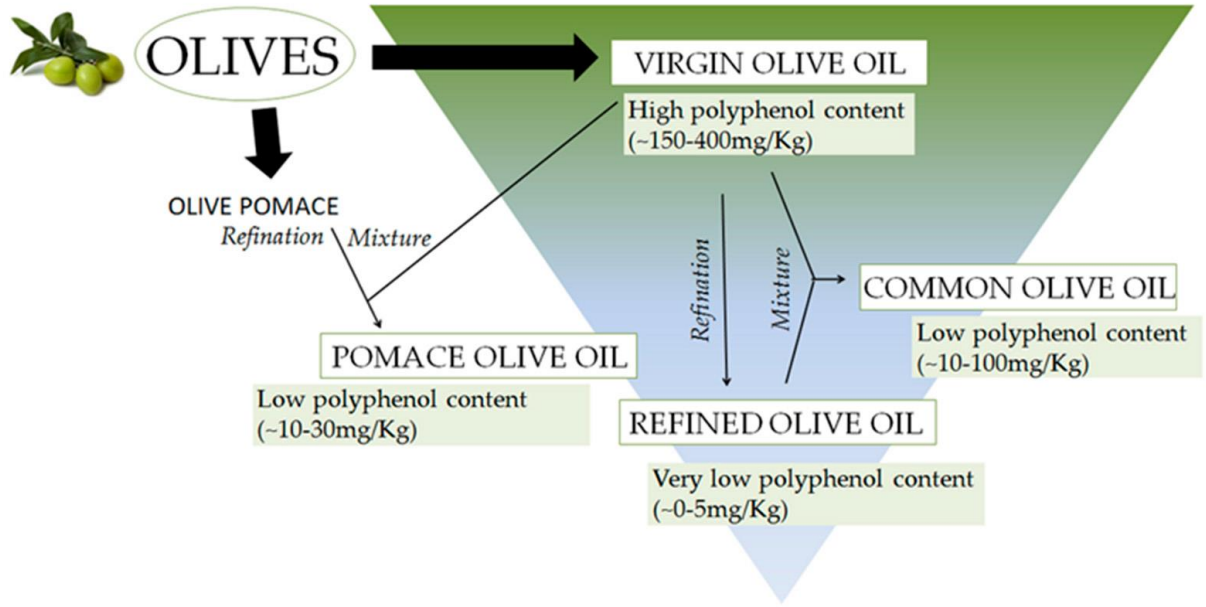

Figure 1. The concentration of polyphenols in various types of olive oil varies according to the technological process of oil extraction. (Gorzynik-Debicka et al.,2018).

The total phenol content of olive oil may be used to classify it as mild, medium, or robust. total phenol levels in robust olive oils exceed 300 mg/kg, while levels in mild olive oils fall below 180
mg/kg. As a result, oil analysis that measures total Phenol provides manufacturers with guidelines for labelling their oil (Visioli et al, 1998). Egypt is one of the largest producers and consumers of olive oil. However, there has been no report, to our knowledge, on the total polyphenolic antioxidants concentrations in Egyptian olive oil. The primary objective of this study was to determine the total polyphenolic antioxidants concentrations of olive oil consumed in Egypt since olive oil is an essential component in our daily life.

2. Methodology

2.1 Chemicals and reagents

Sigma provided the gallic acid and folin ciocalteu reagent (St. Louis, MO, USA). All of the additional chemicals were of analytical grade.

Determination of total phenolic compounds (TPC) in olive oil

The concentrations of TPC in all samples were estimated by a UV spectrophotometer (Jenway-UV-VIS Spectrophotometer) as characterised by (Škerget et al.,2005). The used reagent was Folin-Ciocalteu reagent (AOAS, 1990**)**. To 1 mL of diluted olive oil (1 g in 5 mL ethyl acetate), 2.5 mL of Folin-Ciocalteu reagent 10% , and 2 mL of Na_2CO_3 (7.5 %) were added. the samples were incubated at 50 degrees Celsius for 5 minutes and

then samples were cooled. At 765 nm, the absorbance was measured. TPC was calculated as gallic acid equivalent (GAE) using the calibration curve and the following linear equation:

$$y=0.0203x+0.1194$$
$$R^2 = 0.9987$$

Where *y* represents absorbance and *x* represents concentration (mg GAE g^{-1} extract)
R^2 represents Correlation Coefficient.

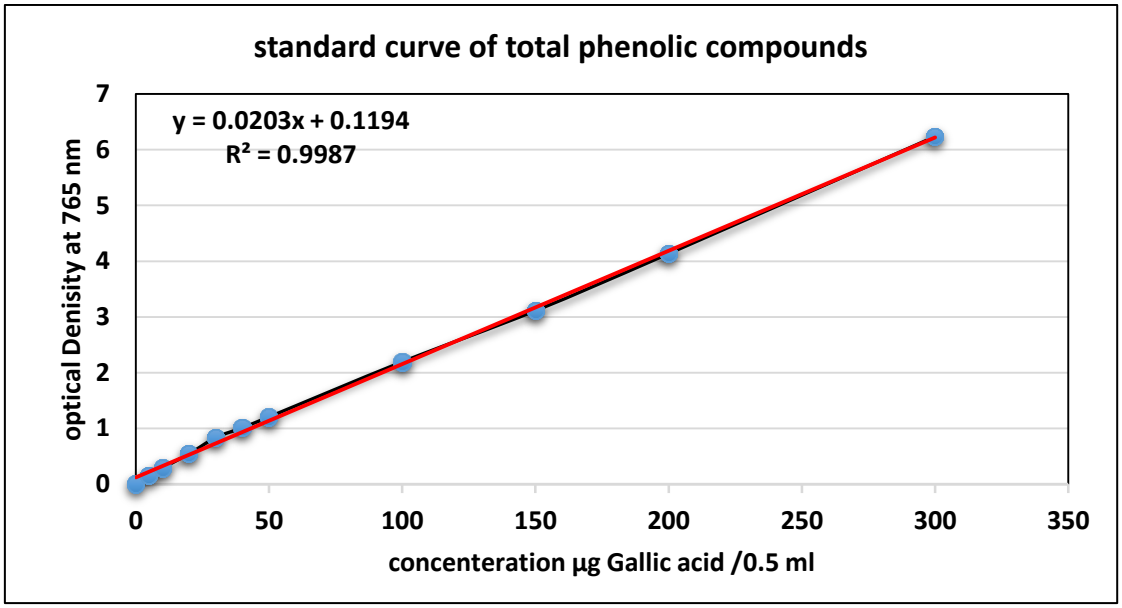

Figure 2: Calibration curve of TPC using gallic acid

2.2 Preparation of phenolics standard curve

After dissolving 10 mg of gallic acid in 10 mL of ethyl acetate, 0, 5, 10,20, 30, 40, 50, 100, 150, 200, and 300 µL were transmitted into test tubes and the volume of each test tube was filled to 500 µL with ethyl acetate. To 0.5 mL of diluted gallic acid, 2.5 mL of Folin-Ciocalteu reagent (diluted 1:10 with distilled water) and 2 mL of Na_2CO_3 (7.5 %) were added. For a blank 0.5 mL of ethyl acetate was used. the samples were incubated at 50 degrees Celsius for 5 minutes and then samples were cooled. At 765 nm, the absorbance was measured **(Table 1)**.

Table 1: Absorbance at various concentrations 765 nm gallic acid standards

Concentration (µg gallic acid/500µL)	Optical density at 765 nm
0	0
5	0.15
10	0.29
20	0.544
30	0.835
40	1.012
50	1.206
100	2.194
150	3.112
200	4.135
300	6.236

3. Results and Discussion

Table 2: The total polyphenol concentration in olive oil sample expressed as ppm of Gallic acid

Samples	total concentration of polyphenol (ppm)
1	33.23
2	0.00
3	251.87
4	141.44
5	33.48
6	237.75
7	0.00
8	285.70
9	281.02
10	111.40
11	109.75
12	0.00
13	247.36
14	123.22
Control	0.00

14 samples of olive oil were collected from different locations in Saini and some were purchased from supermarkets. As shown in table 2, there are variations in concentrations of total polyphenolic in the analyzed olive oil samples. The variation of total polyphenolic concentrations may be due to a variety of considerations including soil, irrigation, environment, level of ripeness, and production technologies, all of which can have a massive effect on an olive oil's total polyphenol content (Baiano et al., 2013) . Furthermore, as polyphenols slow oxidation in the oil, their levels will slowly reduce during storage. Considering such inevitable losses, an extremely high polyphenol concentration is required to ensure a longer shelf life and improved health characteristics. (Houshia et al.,2014). Adulteration of olive oil is a global issue that could be common; additionally, oils sold as EVOO frequently fail to satisfy the grade of minimum quality standard . Two-thirds of imported commodity olive oils classified "extra virgin" failed federal degree standards , according to a 2010 UC Davis Olive Center study. The health consequences of EVOO are highly related to its phenol concentration, so it's very important to investigate the total polyphenol content. It is worth noting that this is the first time determining the total contents of polyphenol antioxidants in some Egyptian virgin and extra virgin olive oil.

4. Conclusion

The concentrations of of total phenolic compounds in 14 samples of virgin and extra virgin were measured by a UV spectrophotometer. The total amount of phenolics in olive oil samples may vary due to crop species, origin, agricultural techniques, fruit maturation, potential infestation by the olive fly Bactrocera oleae, extraction techniques, and storage conditions. The virgin and extra virgin olive oil contains total phenolic content in the range of 150 to 400 mg/kg. It was found that most of concentrations of the analyzed virgin and extra virgin olive oil samples fall within this range of higher total phenolic content. It was found that few numbers of analyzed olive oil samples contain low concentrations of polyphenol content and this reduction in the polyphenol content may be due to cheating with refined olive oil or mixing virgin olive oil with refined olive oil. Since the health benefits of virgin or extra virgin olive oil are strongly related to phenol content we recommend there should be quality control on samples with low polyphenol content that are labeled and sold as virgin or extra virgin olive oil.

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