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Effect of cultivar and geographical area on sterols and quality indices of olive oil

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Abstract
The aim of this study examined the effects of three different areas (Siwie, Ben suef and k64 Cairo Alexandria desert road) that have important differences in climatic and pedologic conditions on the quality indices (free fatty acid, peroxide value and K_{232}, K_{268}), oxidative stability, total phenolic compounds, o-diphenol, fatty acids and sterols composition of extracted oils from three common cultivars (Koronakii, Coratina and Maraqi) growing in Egypt. The obtained results were compared with Standard of International Olive Council.The research results showedthat the quality indices, oxidative stability,fatty acids and sterols composition of the oils were largely influenced by the origin zone, i.e.,by the climatic and pedologic factors of the production environment.

Keywords: Climatic and pedologic condition, olive varieties, sterol, quality indices

1. Introduction

Among many characteristics influencing of the high adult human life expectancy and low rates of chronic disease (including coronary heart disease and some cancers) in the Mediterranean area, a conspicuous factor is olive oil. The important nutraceutical effects of olive oil are due to its balance between oleic, linoleic and linolenic acids, as well as minor components (Krichene *et al*., 2010) [21]. Also, the nutritional value of olive oil is one of its defining characteristics. Having been specified by the International Olive Council (IOC) and the Commission of the European Communities (EEC), the quality of olive oil is determined on the basis of parameters including free fatty acid content, peroxide value, UV absorbency, and other parameters such as fatty acid profile and total unsaponifiable matter, of which sterols are the main constituents (Wiesman, 2009) [33]. The fatty acid profile of olive oil has been used not only for characterization and quality evaluation but also for identification of the growing area (Gomez-Gonzalez *et al*., 2011) [13]. The fatty acid composition of virgin olive oil has great importance from a health point of view. Several studies have shown the dietary importance of fatty acid composition of lipids (Aguilera *et al*., 2000) [3]. Diets rich in monounsaturated fatty acid and lower in saturated fatty acids lowered low-density lipoprotein (LDL) cholesterol and total cholesterol without altering the beneficial high-density lipoprotein cholesterol levels (Matson and Grundy 1985; and Mensink and Katan, 1992) [26, 27]. Aviram and Eias (1993) [7] noted that LDL incubated with oleic acid was less oxidized than others with linoleic and arachidonic acid. Specific profile of sterols in each oily fruit are also used to detect adulteration or to check the genuineness of the olive oil, as it can be considered a distinct "fingerprint" (Lukic *et al*., 2013; and Temime *et al*., 2006) [23, 31].

The international olive council (IOC) imposes limits or ranges for each type of sterol profiles outside of these ranges could suggest that the oil is not genuine. The required sterol profile (as % of total sterols) is as follows: cholesterol ≤ 0.5%, brassicasterol ≤ 0.1%, campesterol ≤ 4.0%, stigmasterol ≤ campesterol in edible oils, Δ-7-stigmasterol ≤ 0.5% β-sitosterol + Δ-5-avenasterol+Δ-5-23-stigmastadienol + clerosterol + sitostanol + Δ-5-24-stigmastadienal ≥93.00% (International olive Council, 2009).

These parameters are influenced by a wide variety of factors such as olive cultivar (Hashempour *et al*., 2010; Mailar *et al*., 2010; and Manti-Djebali *et al*., 2012) [17, 24, 25], ripening stage(Bangana *et al*., 2013) [8]. And growing area (Aguilera *et al*., 2005) [2]. Among these factors, the olive growing area and cultivar are undoubtedly of primary importance (Temime *et al*., 2008) [32], and each combination of cultivar and cultivation zone has a different chemical composition. Thus, interest in the effect of these parameters on the oil´s quantity and quality has been increasing (Krichene *et al*., 2010 and Mailer, 2010) [21]. The relationships between analytical oil variables and pedologic and climatic variables have been less studied (Guerfel *et al*., 2009, and Lazzez *et al*., 2011) [14, 22]. According to Guerfel *et al*., (2009) [14]. Some sterols, triterpenic alcohols and hydrocarbons are correlated negatively with altitude. The influence of elevation variable on the composition of virgin oils was earlier pointed out (Arslan *et al*., 2013) [6]. observed that the oils from 100m altitude were higher in phenols and unsaturated fatty acids, as well as had higher oxidative stability and free acidity compared to 400 m elevation. The unsaturated fatty acid variable appeared also correlated with low temperatures of the cultivation location (Krichene *et al*., 2010) [21]. The positive influence of low temperatures on the oleic acid and unsaturated fatty acids/saturated fatty acids ratio values was also (Issaoui *et al*., 2010) [20]. Ouni *et al*., (2012) [25, 29] showed that rainfall was correlated negatively with the total oil phenol content and positively with total volatiles.

The objective of this study were to examine the influence of cultivars (Koronakii, Coratina and Maraqi) and growing areas (Siwie, Beni Suef and K64 Giza Governorate) on some qualities (acidity, peroxide value, UV spectroscopy) and oxidative stability, fatty acids and sterols of olive oil extracted.

Materials and methods

Sampling: Healthy olive fruits (Oleaeuropaea, Koronakii, Coratina and Maraqi cultivars), without any kind of infection or physical damage, were picked by using brushes during 2013 season at three different growing areas in Egypt: Siwie, Beni Suef and K64 Giza Governorate. These areas were selected based on their climatic and pedologic characteristics (Table 1). Climatic and pedologic data were obtained from the Egyptian Meteorological Organization.

Reagents and standards

Analytical grade solvents and reagents were used to perform analysis except high performance liquid chromatography (HPLC) eluents that were of HPLC grade and purchased from Fluka (Buchs, Switzerland) and Sigma-Aldrich Chemical (Sternheim, Germany.

Physical characteristics

One hundred fruits from each olive sample were weighed using an electric balance with 0.001 g sensitivity and the average weight of the fruit was calculated:

Moisture content

Moisture content was determined by drying the flesh in an oven at 105 ºC until a constant weight (A.O.A.C.2012).

Oil content

Oil content: was determined a Soxhlet apparatus with hexane (60 -80 ºC b.p), as described by (A.O.A.C.2012).

Oil extraction

On the same day of harvest, the fruits were deleafed, then washed to eliminate any foreign material and extracted olive oil using the continuous extraction system two-phases. The oil percent was calculated on fresh weigh basis directly after oil extraction. Then, oil was extracted by centrifugation, transferred into dark bottles without headspace, and stored in the – 20 ºC freezer until analysis.

Quality indices

The quality parameters of olive oil, such as titratable acidity, peroxide value and K_{232} and K_{270} extinction coefficients, were determined according to the IOC (2009) [18]. Peroxide value, being a crude indicator of the amount of primary oxidation product, is a measure of the active oxygen content. Titratable acidity, being a measurement of hydrolysis breakdown of fatty acid chains is usually expressed percent (%) free fatty acids on the basis of oleic acid, because that is the predominate fatty acid in olive oil. Ultraviolet absorption, a more delicate indicator of oxidation is related to the presence of conjugated dienes and trienes systems (Ultraviolet absorbance at 232 and 270 nm, respectively), are conventionally indicated by K [K_{232} and indication of conjugated polyunsaturated fatty acids in olive oil and K_{270} an indication of carbonylic compounds (aldehydes and Ketones)].

Oil stability

The oxidative stability was estimated using Rancimat 679 apparatus (Metrohm, Herisau Switzerland), by measuring the oxidation induction time. A stream of filtered and dried air (20L/h) was bubbled through the oil samples (5g) contained in reaction vessels, these vessels were placed in an electric heating block, which was set at 100 ºC±2 ºC. The volatile oxidation products were collected in water whose conductivity was continuously recorded. The time taken to reach a fixed level of conductivity was recorded (Gutierrez, 1989) [16].

Total phenolic content

Total phenol and O-diphenol contents were quantified calorimetrically. Phenolic compounds were isolated by triple extraction of a solution of oil (10g) in n-hexane (20ml) with 30 ml of a methanol-water mixture (60:40, v/v). The Folin-Ciocalteau reagent (Merck Schuchardt OHG, Hohenbtumm, Germany) was added to a suitable aliquot of the combined extracts and the absorption of the solution at 725 nm was measured. Values are given as milligrams of caffeic acid per kilogram of oil (Gutfinger, 1981) [15]**.** Ortho-diphenols were also measured calorimetrically at 370 nm after adding 5% (w/v) sodium molybdate in 50% ethanol to the extract (Gutfinger, 1981) [15]. Results are given as milligrams of caffeic acid per kilogram of oil.

Gas chromatography analysis

Methylation of fatty acids

An aliquot of fatty acids, about 10 mg, was dissolved in 2ml hexane and then 0.4 ml of 2N KOH in anhydrous methanol was added (Cossignani *et al*., 2005) [10]. After 3 min, 3 ml water was added. The organic layer, separated by centrifugation, was dried over anhydrous sodium sulfate, and then concentrated, with a N2 stream to around 0.5 ml for GC analysis of fatty acids methyl esters (FAME) as described below.

GC analysis of FAME

Agilent 6890 series GC apparatus provided with a DB-23 column (60 m 0.32 mm 0.25 µm) was used. Oven temperatures were 150ºC ramped to 195ºC at 5ºC min -1, ramped to 220ºC at 10ºC min -1 and flow rate was 1.5 min -1. Fatty acids results after the previous procedures steps were transformed into methyl esters and directly injected into the GC.

Sterol analysis

The sterol content was determined according to the official method of Regulation EEC/2568/91(1991) [12]. The oil sample was saponified with an ethanolic potassium hydroxide solution. The unsaponifiable fraction was removed with ethyl ether. The sterols were isolated and analyzed using a Varian CP- 3800 gas chromatograph, equipped with a capillary column J & W Scientific, DB-S (30 m length x 0.25 mm i. d.) coated with a 0.25 µm film thickness, coupled with an FID detector (Varian, Middelburg, The Netherlands). The analytical conditions were as follows: carrier gas, helium: 1.3ml/min flow rate; injector temperature, 280ºC; detector temperature, 300ºC; oven temperature, 270ºC; injection volume; 1µl. The sterols in the non-saponifiable fraction of olive oil were identified by the comparison of retention time with those obtained for corresponding standards. Quantification was made by the addition of an internal standard (α-cholestanol).

Data analysis

At least three replications for each oil sample were performed with each test. The averages and standard deviation were calculated by statistical analysis using SPSS program 10.0 (IBM Corporation, Armonk, Ny). The differences were considered level significant when $P < 0.05$ at a confident level of 95%.

Arrangement of data for statistical analysis was performed by using Microsoft Office Excel (2007).

Results and discussion

Chemical composition

Table 4 shows some chemical composition of the studied cultivar. Koronakii had the lowest oil content expressed as percentage of dry matter in all areas (44.60% in K64, 46.34%in Siwie and 47.30%in Beni suef). Olive cultivars are classified in two group varieties that present a high yield include Maraqi and varieties that percent a middle yield include Coratina and Koronakii. All cultivars grown in K64 showed lower oil content than those grown in Beni suef and Siwie. This could be because of higher temperatures in the Beni suef and Siwie areas. On the other hand, the highest moisture content (57.32%) recorded was that of Maraqi in Beni suef while Maraqi K64 and Siwie showed the lowest value for the moisture content being only (56.13% and 56.99%, respectively).

Quality indices

The quality parameters evaluated in the olive oil samples from the Koronakii, Coratina and Maraqi. Olive oil of different growing area (Beni suef, K64 and Siwie) in Egypt is shown in Table 5. Results indicated the growing area had a significant effect on the free fatty acid content; as seem from Table 5, all cultivars grown in Siwie had higher free fatty acid contents than those grown in Beni suef and k64. This is in good agreement with the findings of Issaoui *et al*., (2010) [20]. That oil produced from cultivars grown in south Tunisia exhibited a higher percentage of acidity than those grown in the north. All studied cultivars had lower peroxide values than the EEC limited of 20 meq.O_2/kg oil (ranging from 2.90 -7.88 meq.O_2/kg oil) for extra virgin olive oil. There were statistically significant differences between the peroxide values of the studied cultivars in each area: the mean peroxide values for the cultivars grown in siwie were higher than for those grown in Beni suef and K64.

All studied samples had low K_{232} and K_{270} values, consistent with the estimated limit for extra virgin olive oils (Table 5). Although the profile of the extinction coefficients at 232 and 270nm showed same slight differences, they were not useful for discriminating between oil samples.

Total polyphenols

Virgin olive oil contains phenolic compounds which affect its stability and flavor. The highest content of total phenols was (230.00 mg/kg) for Koronakii oil grown in Siwie and lowest content of total polyphenols was (115.50 mg/kg) for Coratina oil grown in K64.O-diohenols can be identified as the main chemicals contributing to the overall antioxidant activity of extra virgin olive oils and may therefore play a major role in the preservation of the oils and influencing their organoleptic characteristics (Ouni *et al*., 2012) [25, 29]**.** Direct correlation was observed between total polyphenol, o-diphenol and oxidative stability by Rancimat method (Table 5). Koronakii oil grown in Siwie had the highest total polyphenol and o-diphenol, showed the highest oxidative stability. On the other hand Coratina oil grown in K64 which had the lowest total polyphenols and o-diphenol showed the lowest oxidative stability.

Oxidative stability Oxidative stability is a main parameter in the evaluation of extra virgin olive oil quality, as it gives a reliable estimation of the susceptibility to oxidative degeneration, the main cause of its deterioration in quality (Zarrouk *et al*., 2008) [34]. The oxidative stability determined with Rancimat equipment (Table 5) showed significant differences according to cultivar and growing area. The induction period of samples ranged from 22.40h for Coratina oil grown in K64 to 41.00h Koronakii oil grown in Siwie. Oxidative stability of virgin olive oil depends strongly on the olive cultivar, to the degree that regardless of the growing area, the Coratinacultivar showed the lowest stability. Other reports have described the monounsaturated/polyunsaturated ratio as the main responsible factor for virgin olive oil oxidative stability (Aguilera *et al*., 2005) [2]. Therefore, the low oleic acid content and high linoleic acid content showed by the Coratina cultivars may contribute to the lower oxidative stability, as this causes a low ratio of oleic/linoleic fatty acids (Table 5). Moreover, significant differences were observed in oxidative stability in relation to growing area. Most samples from Siwieshowed higher oxidative stability than those from Beni Suef and K64.

Fatty acid composition

As shown in Table 6 fatty acids were detected in the studied extra virgin olive oils. In general, the distribution of most of the fatty acid composition covered the normal ranges indicated by IOC regulations (2013) [19]. With a minor exception that could have been due to the harvest year or genetic factor (Moghaddam *et al*., 2012) [28]. Palmitic oleic and linoleic acids were predominant in the studied olive oils; the other fatty acids occurred in small amounts. Fatty acid composition may differ from sample to sample. As shown in Table 6 regardless of the growing area, oils extracted from the Coratina and Maraqi cultivars are classified as having high linoleic acid (14.73% and 11.67%) in siwie, respectively and low oleic acid (69.94% and 71.31%) in Siwie, respectively.

Fatty acid profile was significantly affected by growing area for almost all cultivars. In all cases, oleic acid was higher and linoleic acid was lower in the Beni suef and K64 region, which has a higher altitude and higher temperature and lower relative humidity and lower latitude. These results are also in good agreement with those reported by Ranilli *et al*., (1997) [30], in which the percentage of oleic acid is negatively correlated with the relative humidity of the atmosphere.

The percentages of saturated fatty acids (SFA), monounsaturated fatty acids (MUFA) and polyunsaturated fatty acids (PUFA) and the ratio of oleic/linoleic for all samples were also evaluated. Total SFA varied by cultivar for all growing areas ranging between (14.62 and 18.50 %), MUFA (66.25 and 81.77%) and PUFA (7.78 and 15.81%), respectively, some authors have used the total fatty acid composition to distinguish olive oils from Greece, Italy and Tunisia according to the growing area (Guerfel *et al*., 2009**)** [14]**.**

Sterols

The sterols composition of the studied virgin olive oil is listed in Table 7. The β-sitosterol was the predominant phytosterol of all studied cultivars. The highest mean value of β-sitosterols was observed Coratina oil (83.31%) in Siwie, whereas Maraqi oil had the lowest (79.66%) in siwa region. The highest phytosterol levels were followed by Δ-5-avenasterol, which was strongly and negatively correlated with β-sitosterol. Coratina oil in Siwie region had the lowest Δ-5-avenasterol (6.78%) and Maraqi oil in k64 region had the highest (10.34%). Lukic *et al*., (2013) [23] reported that the percentage of β-sitosterol generally decreases

with maturation, while Δ-5-avenasterol increases. All the olive oil samples showed low campestrol content, with a global range from 3.00%Coratina oil in Siwie to 2.40% Coratina oil in Beni suef, which is below the maximum allowable levels established by EU regulations(4%) in all of the oils studied, indicating a peculiarity of these cultivars. Significant differences were observed in the campesterol content in relation to the growing area. Stigmasterol is the main sterol related to the quality of virgin olive oil. There is a negative correlation between its content and oil quality. The mean content in the samples in this study ranged from 0.48% for Koronakii oil growing in Beni suef to 1.09% for Coratina oil grown in siwie; in other words, all cultivars had a very low content, and their amounts were less than the campesterol percentages specified by EU regulations. It is noteworthy that the campesterol/stigmasterol ratio, previously reported as a quality index of olive oils, was higher in the Koronakii oil growing in Beni suef. This is in good agreement with other published results (Krichene *et al*., 2010 and Lukic *et al*., 2013) [21, 23]. that have reported a strong correlation between campesterol content and both cultivar and origin; these studied have stated that in some samples, such as Greek Koronakii, the sterol composition was characterized by a high campesterol level, and it almost always exceeded the legal maximum of 4% established by EU regulations. Apparent β-sitosterol contents, expressed as the sum of the contents of β-sitosterol and other sterols compounds (sitosterol, Δ-5, 24-stigmasterol, clerosterol and Δ-5-avenasterol) formed by its degradation, were generally higher than 93%, the minimum level fixed by EU regulations, thereby proving the authenticity of the corresponding oils except for Maraqi grown in Beni suef and Siwie (94.35% and 93.63%), respectively). Δ-7stigmasterol content in Coratina grown in Beni suef and Coratina grown in Siwie were higher than the legal maximum of 0.5%. Lukic *et al*., (2013) [23]. Expressed that, in some samples, high levels of stigmasterol and its derivatives conditioned the apparent of β-sitosterol not to reach the legal minimum of 93%. The apparent β-sitosterol is significantly affected by growing area. All of the samples analyzed contained more than the established limit of 1.000 mg/kg for total sterols. There significant differences between growing areas ($P \leq 0.05$) for each analyzed samples regardless of the cultivar, from the lowest level in Siwie oils to the highest in K64 oils. These results are consistent with Temime *et al*., (2008) [32]. Who showed the existence of differences according to area?

Generally, many studies reported that some factors effected on sterols content in olive oil such as varieties, topography, geographical areas, soil type and harvest time. The olive variety has an obvious effect on sterols levels. The oil from Maraqi variety showed the lowest percentage of total β-sitosterol (94.35 and 93.63%) growing in B.S and Siwie area under study. While, other varieties were higher in levels total β-sitosterol. This is because the percentage of unsaponifiable matter in the Coratina variety is less than that in other varieties in comparison, the fruit of Koronakii variety in thinner with lower water content. This means that the olive stone containing higher level of sterols other than the olive flesh comprises (Abu-Alruz, *et al*., 2011) [1]. On the other hand the geographical areas was significant effect of sterols levels this may be related to the different climate conditions at each growing site, including rain fall, temperature and humidity. In contrary, sterols levels affected by soil type, the sterols levels was higher in olive oil obtained from olives grown on sandy clay soil. This may be sandy soil reflects the sunand heat thus reduces the evaporation of the water from the soil, while the sandy clay does not.

Table 1: Climatic conditions of production areas.

Parameters	Beni suef (BS)	Siwie (S)	(K 64) Cairo Alexandria desert road
Latitude	29º4´N.	29º12´N	30º01´N
Longitude	31º5´E	25º29´E	31º13´E
Altitude /elevation (m)	29	13	21
Average of maximum temperature	29.30	29.8	28.14
Average of minimum temperature	12.30	13.60	14.28
Average mean temperature	21.8	21.70	21.21
Total of precipitation (rainfall)mm	6.00	9.00	17.00
Average of relative humidity	50.10	45.3	56
Total of sunshine (h/month)	61.30	61.10	34.51

Table 2: Physical and chemical characteristics of the tested soil sample analysis collected from the experimental area

Prosperities	Experimental area		
	Siwie	B.S	k64
Depth	0-50	0-50	0-50
Sand (%)	92.80	37.50	72.50
Silt (%)	2.00	28.00	13.20
Clay (%)	5.20	34.50	14.80
Texture type	Sandy	Sand clay	Loam clay
PH	8.40	7.40	8.00
EC. *	0.12	8.90	10.50
TSS (ppm) **	76.00	5696	6720
Organic matter	0.07	0.3	0.25
Calcium carbonate	9.40	17.00	1.7
Nitrogen (ppm)	160.00	15.00	60.30
Phosphor (ppm)	6.50	0.70	1.26
Potassium (ppm)	140.50	313.00	137.60

EC= Electrical conductivity of soil; TSS= Total soluble solid of soil.

Table 3: Characteristics of the tested water analysis collected from the experimental area.

Prosperities	Experimental area		
	Siwie	B.S	k64
EC mmbes*	2.12	0.55	4.40
TSS ppm **	1356	350	2816
pH	7.17	7.00	7.70

EC= Electrical conductivity of water; TSS= Total soluble solid of water.

Table 4: Chemical composition of olive fruits of some verities

Varieties	Parameters	Growing area		
		B Suef	K64	Siwie
Koronakii	Moisture (%)	56.58±3.12	54.21±4.73	55.15±3.44
	Oil (% w/w)	19.97±1.04	18.20±1.00	18.50±1.09
	Oil (% dry matter)	47.30±2.95	44.60±2.55	46.34±2.11
Coratina	Moisture (%)	58.19±2.55	59.13±3.91	57.21±3.08
	Oil (% w/w)	21.53±1.98	22.32±1.16	32.00±2.11
	Oil (% dry matter)	48.80±3.00	48.65±2.76	48.89±2.66
Maraqi	Moisture (%)	58.23±3.88	56.13±3.23	56.99±3.59
	Oil (% w/w)	18.83±0.98	23.16±1.65	19.00±0.98
	Oil (% dry matter)	50.77±2.66	50.99±3.94	52.78±2.99

Mean value ± standard deviation (SD).

Table 5: Some physicochemical properties of olive oil extracted from three varieties from tree different growing area.

Varieties	Parameters						
	Free Fatty acids (%)	Peroxide value	Absorbance at 232 nm	Absorbance at 268 nm	Oxidative stability (hr)	Total phenolic (mg/kg)	O-diphenol (mg/kg)
CBS	0.23±0.001	2.90±0.61	0.812±0.10	0.18±0.001	24.30±2.87	125.30±7.80	65.20±3.55
C64	0.30±0.12	5.39±0.83	1.210±0.11	0.25±0.001	22.40±2.32	115.50±6.30	60.30±3.21
CS	0.28±0.010	7.62±0.88	0.718±0.01	0.15±0.001	33.40±3.75	166.50±9.55	73.40±4.40
KBS	0.26±0.001	5.14±0.76	0.544±0.01	0.11±0.001	34.50±3.00	183.11±10.20	77.20±5.11
K64	0.31±0.10	5.35±0.61	0.600±0.01	0.09±0.001	39.00±4.11	205.17±11.11	90.30±6.00
KS	0.36±0.010	5.46±0.63	0.531±0.01	0.10±0.001	41.00±4.50	230.00±13.22	110.50±6.99
MBS	0.24±0.01	5.54±0.58	0.566±0.01	0.10±0.001	37.20±3.43	198.20±10.90	85.90±5.87
M64	0.63±0.15	5.50±0.49	0.620±0.01	0.12±0.001	31.85±2.96	153.40±8.84	70.20±4.67
MS	0.48±0.16	7.88±0.88	1.00±0.15	0.22±0.001	25.57±2.78	130.80±7.01	69.50±4/34

Mean value ± standard deviation (SD).

Table 6: Fatty acids composition of olive oil extracted from three varieties from tree different growing area.

Fatty acids	Varieties								
	CBS	C64	CS	KBS	K64	KS	MBS	M64	MS
$C_{16:0}$	11.88±0.88	14.24±0.91	14.59±0.84	14.62±0.96	15.06±0.98	15.26±0.89	11.22±0.66	11.46±0.75	11.99±0.81
$C_{16:1}$	0.55±0.15	0.57±0.13	0.81±0.56	1.37±0.62	1.43±0.65	1.50±0.76	0.61±0.21	0.51±0.15	0.52±0.14
$C_{17:0}$	0.08±0.001	0.07±0.001	0.03±0.001	0.04±0.001	0.05±0.001	0.04±0.001	0.03±0.001	0.05±0.001	0.05±0.001
$C_{17:1}$	0.06±0.001	0.06±0.001	0.06±0.001	0.07±0.001	0.08±0.001	0.07±0.001	0.07±0.001	0.06±0.001	0.07±0.001
$C_{18:0}$	2.60±0.21	2.16±0.19	2.78±0.17	2.84±0.20	2.65±0.22	2.47±0.18	2.89±0.17	2.55±0.16	2.48±0.18
$C_{18:1}$	74.95±6.95	71.65±5.98	64.94±5.00	70.48±6.86	71.84±6.74	70.27±5.99	75.76±7.84	74.75±7.83	71.31±6.57
$C_{18:2}$	8.04±0.88	9.35±0.91	14.73±1.02	8.65±0.87	6.79±0.76	8.40±0.82	7.44±0.63	8.99±0.86	11.67±1.13
$C_{18:3}$	0.73±0.14	0.82±0.15	1.08±0.20	0.85±0.19	0.99±0.22	0.96±0.18	0.82±0.12	0.69±0.10	0.94±0.20
$C_{20:0}$	0.55±0.01	0.47±0.09	0.44±0.01	0.59±o,11	0.59±0.11	0.49±0.08	0.53±0.07	0.46±0.10	0.46±0.07
$C_{20:1}$	0.44±0.10	0.47±0.09	0.44±0.10	0.35±0.10	0.36±0.10	0.30±0.10	0.45±0.09	0.38±0.09	0.46±0.09
$C_{22:0}$	0.12±0.001	0.14±0.001	0.10±0.001	0.14±0.001	0.16±0.001	0.14±0.001	0.10±0.001	0.10±0.001	0.09±0.001
SFA	15.23±1.03	17.08±1.19	17.94±1.15	18.23±1.17	18.51±1.09	18.50±1.06	14.77±0.98	14.62±0.68	15.07±0.88
USFA	84.77±7.96	82.92±7.99	82.06±8.35	81.15±7.98	81.49±8.11	81.50±8.01	85.15±7.19	85.38±9.01	84.89±8.66
MUSFA	76.00±7.11	72.75±6.09	66.25±5.83	81.77±8.00	73.71±6.88	72.07±6.56	76.80±7.03	75.70±7.11	72.36±6.19
PUSFA	8.77±0.82	10.17±1.00	15.81±1.34	9.50±0.87	7.78±0.77	9.36±0.81	8.26±0.79	9.68±0.98	12.61±1.19
$C_{18:1}/C_{18:2}$	9.32±0.89	7.66±0.69	4.40±0.44	8.14±0.86	10.58±0.95	8.36±0.78	10.18±1.00	8.31±0.79	6.11±0.57

Mean value ± standard deviation (SD).

Table 7: Sterols composition of olive oil extracted from three varieties from tree different growing area.

Varieties	Sterols compounds											
	Cholesterol	Brassicasterol	Campesterol	Stigmasterol	Clerosterol	Δ-7-Campesterol	B-sitosterol	Δ-5-Avenasterol	Δ--5,24-Stigmastadienol	Δ-7-Stigmasterol	Δ-7-Avenasterol	Total β-Sitosterol
CBS	0.23±.001	0.12±0.001	2.40±0.15	0.90±0.14	0.79±0.17	0.45±0.09	80.09±7.98	10.03±1.70	2.34±0. 01	0.52±0.01	0.71±0.16	94.50±7.88
C64	0.42±0.010	0.15±0.001	2.70±0.13	1.00±0.010	0.88±0.18	0.53±0.10	82.89±8.01	8.80±1.00	1.43±0.01	0.51±0.01	0.84±0.15	95.42±7.41
CS	0.40±0.11	0.16±0.001	3.00±0.19	1.09±0.18	0.91±0.21	0.56±0.10	83.31±7.89	6.78±1.13	1.47±0.10	0.56±0.08	0.89±0.1 1	93.99±6.66
KBS	0.32±0.15	0.15±0.001	2.89±0.18	0.48±0.15	0.85±0.16	0.51±0.10	82.78±8.09	8.76±0.98	2.41±0.10	0.44±0.11	0.56±0.09	95.43±8.01

K64	0.26±0.10	0.18±0.001	2.93±0.20	0.99±0.31	0.83±0.17	0.48±0.11	81.89±7.68	10.00±1.07	2.33±0.09	0.49±0.12	0.80±0.18	96.51±9.11
KS	0.10±0.001	0.10±0.001	2.64±0.18	0.95±0.24	0.80±0.15	0.47±0.09	80.90±7.38	8.87±0.97	2.28±0.001	0.41±0.11	0.77±0.21	94.29±7.75
MBS	0.34±0.01	0.13±0.001	2.56±0.19	0.69±0.20	0.77±0.13	0.38±0.11	79.98±6.99	9.74±1.19	2.48±0.13	0.43±0.12	0.57±0.07	93.75±7.00
M64	0.22±0.001	0.11±0.001	2.72±0.18	0.80±0.19	0.80±0.18	0.49±0.12	81.45±7.17	10.34±1.83	2.26±0.001	0.49±0.10	0.67±0.22	96.07±8.74
MS	0.18±0.001	0.14±0.001	2.90±0.20	0.78±0.20	0.78±0.15	0.52±0.12	79.66±7.04	9.41±1.11	2.30±0.001	0.42±0.11	0.54±0.12	93.17±7.23

Mean value ± standard deviation (SD).

Conclusion

In the end, it can be summed up to say that there are a lot of factors that affect the (oil content and quality attributes, fatty acids, sterol content) in the oil extracted from (Coratina, Koronakii and Maraqi varieties) cultivated in three regions in Egypt. The oleic acid was higher and linoleic acid was lower in the Beni suef and K64 region. Δ-7-stigmasterol content in Coratina grown in Beni suef and Coratina grown in Siwie were higher than the legal maximum of 0.5%. The highest content of total phenols was (230.00 mg/kg) for Koronakii oil grown in Siwie and lowest content of total polyphenols was (115.50 mg/kg) for Coratina oil grown in K64. The induction period of samples ranged from 22.40h for Coratina oil grown in K64 to 41.00h Koronakii oil grown in Siwie.

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References

1. Abu-Alruz K, Afaneh IA, Quasem JM, Hamiat MA, Abbady J, Mazahreh AS. Factors affecting D-7-stigmasterol in Palestinian olive oil. Journal of Applied Sciences. 2011; 11(5):797-805.
2. Aguilera MP, Beltra´n G, Ortega D, Ferna´ndez A, Jime´nez A, Uceda M, Characterization of virgin olive oil of Italian olive cultivars: 'Frantoio' and 'Leccino', grown in Andalusia. Food Chem. 2005; 89(3):387-391.
3. Aguilera MC, Ramirez-Tortosa MD, Gil A, Do MUFA and PUFA have beneficial effects on development of cardiovascular disease? In Resent Research Development in Lipid (Advances in Lipid Research); Pandalai S. G., Ed.: Transworld Research Network Trivandrum, India, 2000, 369-390.
4. American Oil Chemists' Society Official Methods and Recommended Practices of the American Oil Chemists' Society, 5th edn. AOCS Press, Champaign, 1998.
5. AOAC Official Methods of Analysis of the Association of Official Analytical Chemists, 18thedn (edited by W. Horwitz). Gaithersburg, MO: AOAC, 2012, 62.
6. Arslan D, Karabekir Y, Schreiner M. Variations of phenolic compounds, fatty acids and some qualitative characteristics of Sarıulak olive oil as induced by growing area. Food Res Int 2013; 54(2):1897-1906.
7. Aviram M, Eias K. Dietary olive oil reduces low-density lipoprotein uptake by macrophages and decreases the susceptibility of the lipoprotein to undergo lipid peroxidation. Ann. Nutr. Metab. 1993; 37:75-84.
8. Bengana M, Bakhouche A, Lozano-Sa´nchez J, Amir Y, Youyou A, Segura-Carretero A, Ferna ´ndez-Gutie´rrez A. Influence of olive ripeness on chemical properties and phenolic composition of Chemlali extra-virgin olive oil. Food Res Int 2013; 54(2):1868-1875.
9. Cairo (A) Climate Normals National Oceanic and Atmospheric Administration. Retrieved, 2014, 1961, 1990.
10. Cossiganani L, Simonetti MS, Damiani P. Biocatalyzed acidolysis of olive oil triacylglycerols with 9c, 11t and 10t, 12c isomers of conjugated linoleic acid Eur. Food Res. Technol. 2005; 220:267-271.
11. Covas MI, Ruiz-Gutierrez V, de la Torre R, Kafatos A, Lamuela-Raventos RM. Minor components of olive oil: Evidence to date of health benefits in humans. Nutri. Rev. 2006; 64:20-30.
12. EEC Characteristic of olive and olive pomace oils and their analytical methods. Regulation EEC/2568/91 and later modification. Official Journal of European Communities, 1991; 248:1-82
13. Gomez-Gonza lez S, Ruiz-Jime´nez J, Luque de Castro MD. Oil content and fatty acid profile of spanish cultivars during olive fruit ripening. J Am. Oil Chem. Soc. 2011; 88(11):1737-1745.
14. Guerfel M, Ouni Y, Taamalli A, Boujnah D, Stefanoudaki E, Zarrouk M. Effect of location on virgin olive oils of the two main Tunisian olive cultivars. Eur. J Lipid Sci. Technol. 2009; 111(9):926-932.
15. Gutfinger T. Polyphenols in olive virgin oils. J. Am. Oil Chem. 1981; 58:996-998 .
16. Gutierrez F. Determination of the oxidative stability of virgin olive oils. Comparison of the active oxygen and the Rancimat methods, Grasas Aceites, 1989; 40:1-5.
17. Hashempour A, FotouhiGhazvini R, Bakhshi D, Asadi S. Fatty acids composition and pigments changing of virgin olive oil (Oleaeuropea L.) in five cultivars grown in Iran. Aust. J Crop Sci. 2010; 4(4):258-263.
18. IOC International olive oil council. International trade standard Appling to olive oils and olive- pomace oils.COI/T.15/NC 3/Rev, 2009, 2.
19. IOC Trade standard applying to olive oils and olive pomace oils. COI/T.15/NCNO.3/Rev.7,www.internationaloliveoil.org/documents/viewfile/3615, 2013.
20. Issaoui M, Flamini G, Brahmi F, Dabbou S, Hassine KB, Taamali A, Chehab H, Ellouz M, Zarrouk M, Hammami M. Effect of the growing area conditions on differentiation between Chemlali and Che´toui olive oils. Food Chem. 2010; 119(1):220-225.
21. Kriche`ne D, Allalout A, Salvador MD, Fregapane G, Zarrouk M.).Fatty acids, volatiles, sterols and triterpenic

alcohols of six monovarietal Tunisian virgin olive oils. Eur. J. Lipid Sci. Technol. 2010; 112(3):400-409.
22. Lazzez A, Vichi S, Kammoun NG, Arous MN, Khlif M, Romero A, Cossentini M. A four year study to determine the optimal harvesting period for Tunisian Chemlali olives. Eur. J. Lipid Sci. Technol. 2011; 113(6):796-807.
23. Lukic M, Lukic I, Krapac M, Sladonja B, Pilizota V. Sterols and triterpenediols in olive oil as indicators of variety and degree of ripening. Food Chem, 2013; 136(1):251-258.
24. Mailer RJ, Ayton J, Graham K. The Influence of growing region, cultivar and harvest timing on the diversity of Australian olive oil. J Am Oil Chem. Soc. 2010; 87(8):877 884.
25. Manai-Djebali H, Kriche`ne D, Ouni Y, Gallardo L, Sa´nchez J, Osorio E, *et al*. Chemical profiles of five minor olive oil varieties grown in central Tunisia. J Food Compos Anal. 2012; 27(2):109-119.
26. Matson FM, Grundy SM. Comparison of effects of dietary saturated, monounsaturated a polyunsaturated fatty acids on plasma lipids and lipoproteins in man. J Lipid Res. 1985; 26:194-202.
27. Mensink RP, Katan MB. Effect of dietary fatty acids on serum lipids and lipoproteins. Arteroscler, ThrombVasc. Biol, 1992; 12:911-919.
28. Moghaddam G, Heyden YY, Rbiei Z, Sadeghi N, Oveisi MR, Jannat B *et al*. Characterization of different olive pulp and kernel oils. J. Food Compos. Anal. 2012; 28(1):54-60.
29. Ouni Y, Taamali A, Guerfel M, Abdellly C, Zarrouk M, Flamini G. The phenolic compounds and compositional quality of Chetoui virgin olive oil: Effect of altitude. African, J of Biotech, 2012; 11(5):11842-11850.
30. Ranalli A, De Mattia, G Ferrante, M Giansante L. Incidence of olive cultivation area on the analytical characteristics of the oil.Riv.ItalSostanze Grasse 1997; 74(11):501-508.
31. Temime SB, Campeol E, Cioni PL, Daoud D, Zarrouk M. Volatile compounds from Che´toui olive oil and variations induced by growing area. Food Chem. 2006; 99(2):315-325.
32. Temime S, Manai H, Methenni K, Baccouri B, Abaza L, Daoud D. Sterolic composition of Che´toui virgin olive oil: influence of geographical origin. Food Chem. 2008; 110(2):368-374.
33. Wiesman Z. Desert Olive Oil Cultivation: Advanced Biotechnologies, Academic Press, 2009; 266-288, 299, 36, and 38.
34. Zarrouk W, Haddada FM, Baccouri B, OueslatiI, Taamalli W, Fernandez X, *et al*. Characterization of virgin olive oil from Southern Tunisia. Eur. J. Lipid Sci. Technol. 2008; 110(1):81-88.