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Oxidizability and Fatty Acids Composition in Developing Fruits of High and Low Oil Olive (*Olea europea L.*) Cultivars

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ABSTRACT

The objective of the present study was to Compare of the fatty acids accumulation pattern and oil oxidizability during development and ripening of Mary and Shengeh fruits as high and low oil olive cultivars(of *Olea europaea* L.), respectively, at the same environmental condition. Gas-chromatographic analyses of main fatty acids (palmitate (C16:0), stearate (C18:0), oleate (C18:1), and linoleate (C18:2)) showed a positive relation between stearate pattern and oleate, however, an inverse relationship was observed between palmitate and linoleat. Moreover, oil content was reported to be positively correlated with stearat and oleate and inversely related with palmitate and linoleate. Also, there was positive correlation between linoleate and COX value. Based on the results obtained, it is concluded that, accumulation and composition of major fatty acids are influenced by genotype and for selection of high oil contents olive cultivars with low oxidizability, cultivars with high oleate and stearate, also with low palmitate and linoleate could be considered.

KEYWORDS: Olive development; Gas chromatography; fatty acid profile; Oxidizability

INTRODUCTION

Olea europaea is one of the principal and widespread fruit trees in Mediterranea and has a major economic impact on the production of olive oil (Loumou and Giourga, 2003).The olive oil is commonly used worldwide for its exceptional properties. The unusual fatty acid composition of olive oil has attracted increasing interest(Colomer and Mene´ndez2006).One of the main features of olive oil is having a high oil percentage of oleat (60–80% of total fatty acid content), followed by other constituents, including linoleat, palmitate, stearate and linoleat (C18:3), respectively (Uceda and Hermoso, 1998; Bianco et al., 2013). Oleat plays important role in decreasing the low-density lipoprotein (LDL) cholesterol and increasing the high-density lipoprotein (HDL) cholesterol levels in the blood. Previous studies performed on animal diets suggested that the defensive effect of olive oils against breast cancer is because of their high oleat content (Conde et al., 2008)

It was previously shown that a profitable unsaturated fatty acids/Saturated fatty acids (ΣUSFA/ΣSFA) ratio could be assumed as a useful marker to determine the quality of edible oil (Lee et al., 1998). It was suggested that oils with higher ratios of ΣUSFA/Σ have favorable health benefits (Rabrenovic et al., 2011). Moreover, the calculated oxidizability (COX) value, which is based on unsaturated fatty acid contents in the oils, is a useful factor for measuring the oil's susceptibility to autoxidation (Moghaddam et al., 2012).

The quality of olive oil is mainly affected by the metabolic processes occurring during fruit development. Olive fruit development includes 5 main phases: I) fertilization and fruit set (0–30 DAF[1]), II) seed development (30–60DAF), III) pit hardening (60–90 DAF), IV) mesocarp development and intense oil accumulation (90–150 DAF), and V) ripening (since 150 DAF) (Bianco et al., 2013; Giovannoni, 2004).

To date, several researches have been conducted on the effect of genetic factor on olive oil quality for different olive cultivars (Cerretani et al., 2006; Hashempour et al., 2010).However, little is known about the impact of genetic, olive oil content, and lip genesis process on fatty acid composition of olive oils and COX of Mary and Shengeh cultivars in Iran. Therefore, the aim of this study was to gain knowledge about these factors in olive oil. For this purpose, we selected two olive cultivars, Mary and Shengeh, with high and low oil content respectively.

METHODS AND MATERIALS

Plant material

Mary (high oleat) and Shengeh (low oleat) genotypes of olive from Tarum international olive collection station (Zanjan province) were selected. Fruits samples were harvested 90, 120, 150 and 180 days after flowering (DAF). Then, olives were washed with distilled water and stored at -80 temperatures.

[1]Days after flowering

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Oil extraction

For oil extraction, olive fruits were pressed with a hammer mill. After malaxation, oil separated by 30 min centrifugation of the produced olives at 5000 rpm. Collected oil stored in the dark at 4ºC(Guardia-Rubio et al., 2007).

Gas chromatography (GC) analysis

The fatty acids composition of extracted oils was determined by gas chromatography, according to the methods described in regulation of EEC 2568/91.

Fatty acid methyl esters (FAMEs) were prepared by adding 200 µl saturated methanolic potassium hydroxide solution and then 2 ml n-hexane on 100 µl of oil samples for at last 20 min. GC analysis was performed on an ACME 6100 Younglin GC, by a fused-silica capillary column (60 m × 0.32mm × 0.5 µm film thicknesses, Teknokroma, Barcelona, Spain). Selected temperatures of injector and oven were 240 ^{0}C and 185^{0}C, respectively. Flow rate of helium as the carrier gas was maintainedat 1 mL/min.

Calculated oxidizability value (COX)

The Cox value of the olive oils was calculated based on the proposed formula.

$$COX = \frac{[1(18{:}1\%) + 10.3(18{:}2\%) + 21.6(18{:}3\%)}{100}$$

where18:1%, 18:2% and18:3% related to oleat, linoleat and linolenat, respectively (moghaddam et al., 2012).

Statistical analysis

Analysis of data was done by one-way analysis of variance (ANOVA) by Excel and version 13 of SPSS software. $p<0.05$ was selected as the significant level. All tests were done in three replicates.

RESULTS AND DISCUSSION

In the present study, palmitate, oleat, stearat and linoleat were evaluated as main fatty acids of olive oil.

Fig 1-I elucidates the variation of palmitate% in two Mary and Shengeh cultivars, during fruit development. Influence of DAF, genotype, and their interaction on percentage of palmitate showed that palmitate was only influenced by genotype and higher value of palmitate was demonstrated in Mary. Therefore, no significant variation was observed at 90, 120, 150 and 180 DAF in both cultivars. However, a slightly decrease was observed in the mentioned cultivars, until reaching full maturirty. Stream of variation for palmitate% during olive fruit development in this study was in agreement with the study of Sakouhi (Sakouhi et al.,, 2011).

As it is shown in figure 1-П, stearate had significantly risen during the ripening of Shengeh cultivar from 0.73 ±0.02 to 1.38±0.1, but its variation was not noticeable in Mary. Comparison of the data obtained from these two cultivars showed higher amount of stearat% in Shengeh than in Mary. Therefore, genotype and DAF had remarkable effects on stearat% of olive oil. Also, evaluation of both Fig. 1-I and 1-П revealed that palmitate and stearate have converse accumulation patterns. In fact, palmitate and stearat contents of vegetable oils depend on the activity of thioesterase enzyme (Harwood, 1996).

Furthermore, oleate accumulation pattern was observed to be different in each studied cultivars. Fig. 1-Ш indicated that the percentage of oleate had significantly decreased, during development stage of Shengeh cultivar but remained in variable in Mary. Thus, it is deduced that DAF, genotype, and their interaction could extremely influence on oleate pattern during fruit development.

In the study carried out by Sakouhi et al., the major biosynthetic phase of oleat was reported to occur between 90 and 170 DAF, in which the highest accumulation of oils occur in this period (Sakouhi et al., 2011). Furthermore, the results of the current study also displayed divergent accumulation patterns for palmitate and oleate (Fig. 1-I and 1-Ш). Indeed, conversion of palmitate to oleate is the reason of palmitate decreasing.

Linoleat and oleate illustrated an opposite accumulation patterns in both cultivars (Fig. 1- IV). This increase is due to the activity of oleate desaturase during oil accumulation that alters oleate into linoleate (Gutiérrez et al., 1999; Sanchez and Harwood., 2002).

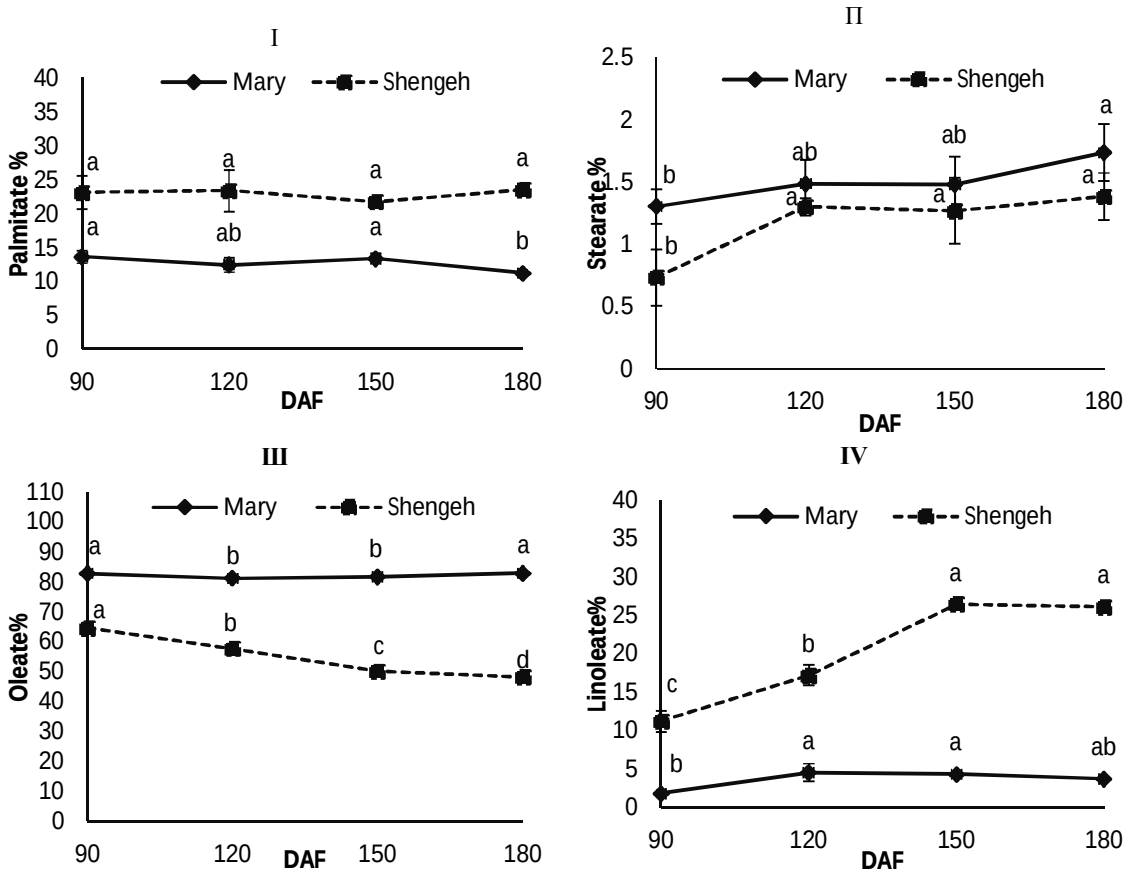

Figure1. Changes occurred in content of the major fatty acids (%) during the maturity stage of Mary and Shengeh olive cultivars. I) Palmitate, II) stearate, III) oleate and IV) linoleate patterns.

Comparison of palmitate, stearat, oleate and linoleate patterns (Fig.1) demonstrated that stearate pattern positively associated with oleate but had an converse relation with palmitate and linoleat. Although oil content had a positive correlation with stearate and oleate, it revealed an inverse relationship with palmitate and linoleate. Results of this study consent with those previous studies reporting a relation between the mentioned fatty acids in sesame (Brar, 1982), sunflower (Flagella et al. 2002), cotton (Liu et al. 2002), and rapeseed (Mo"llers and Schierholt, 2002). Palmitate is a16-carbon fatty acid that after elongation converted to 18-carbon stearate and with subsequent desaturation by desaturase enzymes, altered to oleate and linoleat. A defect in elongation step leads to an increase in the palmitate and decrease in oleate content of oils. In fact, palmitate has a high affinity to oleate, as a favorable substrate (Salas et al. 2000).

Moreover, in the present study the ratio of oleate to linoleate was also observed to be considerably decreased, which might be due to an increase in linoleate synthesis. In general, the simultaneous increase in unsaturated fatty acid and decrease in antioxidant during fruit ripening, make the oils more inclined to oxidation (Rotondi et al., 2004). As can be seen from Table 1, there were different patterns of unsaturated and saturated fatty acids ratio (ΣUSFA/ΣSFA) in the two investigated cultivars, thus during development this ratio was observed to be increased in Mary but decreased in shengeh which may be because of oleate and linoleat accumulation in Mary and Shengeh, respectively.

Table1. Fatty acid composition and COX value of Mary and Shengeh, during fruit development and ripening.

Fatty acid composition%	Codex	Mary				Shengeh			
		90 DAF	120 DAF	150 DAF	180 DAF	90 DAF	120 DAF	150 DAF	180 DAF
Palmitic acid	7.5-20	13.67^{a}	12.47^{a}	13.39^{a}	11.27^{a}	23.13^{a}	23.4^{ab}	21.78^{a}	23.55^{b}
Stearic acid	0.5-5	1.27^{b}	1.49^{ab}	1.48^{ab}	1.74^{a}	0.73^{b}	1.30^{a}	1.27^{a}	1.38^{a}
Oleic acid	55-83	82.86^{a}	81.19^{b}	81.78^{b}	82.98^{a}	64.62^{a}	57.77^{b}	50.21^{c}	47.30^{d}
Linoleic acid	3.5-21	1.83^{b}	4.54^{a}	4.35^{a}	3.63^{a}	11.21^{c}	17.27^{b}	26.43^{a}	26.08^{a}
ΣUFA/ΣSFA		5.68^{a}	6.15^{b}	6.36^{c}	6.68^{d}	3.18^{ab}	3.04^{b}	3.33^{a}	2.99^{b}
Cox value		1.08^{b}	1.33^{a}	1.32^{a}	1.26^{ab}	1.84^{c}	2.40^{b}	3.27^{a}	3.24^{a}

Oxidation index

Table1 shows that oil Cox values raised during fruit development in both cultivars. The amount of Cox values in Shengeh was higher than that of Mary. The maximum and minimum Cox values of oil were found in Shengeh at 180 DAF (3.24) and 90 DAF of Mary (1.08), respectively. On the other hand, as can be seen in Fig.2, there was a highly positive linear correlation between COX values and linoleat pattern in both cultivars. Since poly unsaturation of fatty acid increases the ability of autoxidation, correlation between COX and linoleat is deemed rational.

Furthermore, a negative relation between the ratio of Cox value and ΣUSFA/ΣSFA patterns of both cultivars were observed.

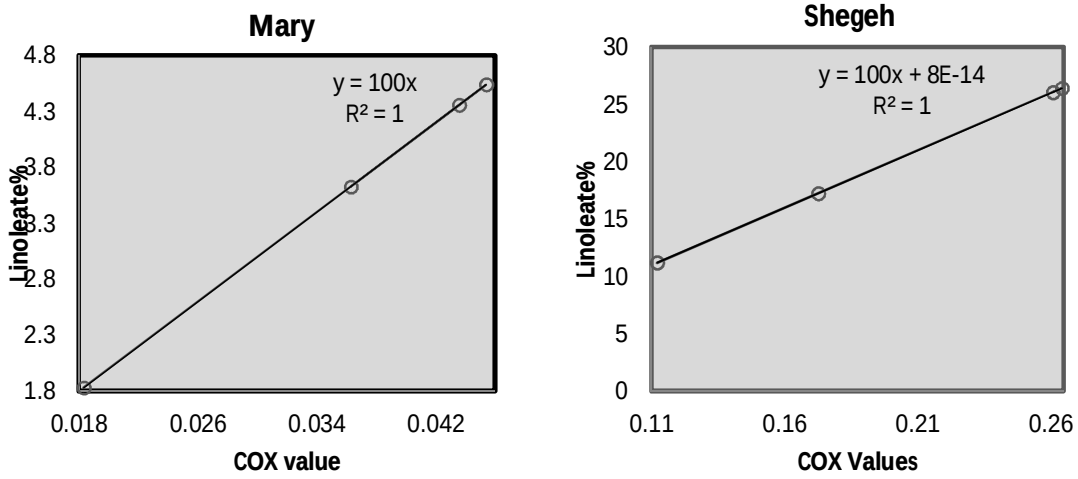

Figure2. Relation between COX values and linoleat pattern in Shengeh and Mary cultivars.

Conclusions

In summary, since environmental conditions were similar for both of cultivars, fatty acid composition of olive cultivars during fruit development might be influenced by genetic factors.

Based on our findings, oil content was shown to be positively associated with stearat and oleate amount with an opposite association with palmitate and linoleate. Also, there was appositive relationship between linoleate and COX value.

It is suggested that for selection of olive cultivars with properties such as increased oil content, high ratio of ΣUSFA/ΣSFA and also low oxidizability, cultivars with high oleate and stearate, as well as low palmitate and

linoleate would be a suitable choice. Therefore, our findings may be profitable for breeding work headed for improving the oil yield of olive.

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