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EFFECT OF SHELF CONDITIONS ON THE PHENOLIC FRACTION AND OXIDATION INDICES OF MONOVARIETAL EXTRA VIRGIN OLIVE OIL FROM CV. 'TAGGIASCA'

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The quality of monovarietal extra virgin olive oil from cv. 'Taggiasca' is influenced by many factors that have impact on shelf-life as well as on sensory and healthy properties of the product. The aim of the work was to recreate the conditions similar to those in consumer sales point (conditions of "shelf"), maintaining the olive oil packaged in dark-green bottles at room temperature (between 18 and 25 °C) under artificial light and away from heat sources, monitoring the oils up to 12 months from bottling with quarterly sampling for the main chemical, physico-chemical, and sensory parameters related to the quality. After one year of storage, an organoleptic alteration with reduction of the attributes 'fruity', 'pungent', and 'bitter', as well as the occurrence of 'rancid' defect, was observed. This alteration was found to be accompanied by a decrease in phenolic substances and tocopherols and an increase in primary and secondary oxidation products. The composition of the volatile fraction showed a slight increase of substances related to rancid defect, a constant trend of compounds related to fruitiness, and a slight decrease in alcohols. It can be concluded that the optimum time of storage of the oil under the above-mentioned conditions is approximately 9 months.

Keywords: monovarietal extra virgin olive oil, *Olea europaea*, rancid defect, shelf-life

Lipid hydrolysis and oxidation are the main causes of the extra virgin olive oil (EVOO) quality deterioration and its reaction rate determines the shelf-life of this product. These processes lead to the formation of off-flavours along with a decrease in nutritional property (Angerosa et al., 2004). EVOO is a source of natural antioxidants, such as tocopherols, chlorophyll, and carotenoid pigments, and biophenols, involved in different mechanisms providing an effective defence against free radicals (Servili et al., 2014). The antioxidant content depends on many factors, in particular on genetic profile of *Olea europaea* cultivars. The study of the chemical and physico-chemical characteristics of monovarietal extra virgin olive oil (MEVOO) is useful in order to establish the storage time to preserve as long as possible the organoleptic and nutritional quality within the limits established for the extra-virgin category declared by the European Union Commission Regulation EEC (1991) and its subsequent modifications.

The oxidative alterations that occur during the conservation of oil have been widely studied (Gomez-Alonso et al., 2007; Samaniego-Sánchez et al., 2012). The influence of temperature, light, metal contents, the presence of pigments and antioxidants, and oxygen partial pressure on the oxidative process was also studied (Gutierrez & Fernandez, 2002; Stefanoudaki et al., 2010; Ayton et al., 2012; Nabil et al., 2012). However, only few works have been carried out to study the shelf-life of monovarietal extra virgin olive oils (Lanza et

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al., 2013b). Several studies demonstrated that the storage conditions, such as containers, temperature, exposure to light and air, influence the packaged oils (Angerosa et al., 1993; Morello et al., 2004; Méndez & Falqué, 2007; Romani et al., 2007). The purpose of the present work was to reproduce conditions similar to those referred to in sales point (conditions of "shelf"), maintaining the olive oil packaged in dark-green bottles at room temperature (between 18 and 25 °C) under artificial light and away from heat sources, monitoring the oils up to 12 months from bottling with quarterly sampling for the main chemical, physico-chemical, and sensory parameters related to the quality.

1. Materials and methods

1.1. Materials and storage conditions

Monovarietal extra virgin olive oils (MEVOO) obtained from *Olea europaea* L. cv 'Taggiasca' were used in this study. The oils were extracted in industrial mill by three-phase continuous system and then packaged in full filled 1 l bottles of dark-green glass. The bottles were maintained at room temperature (between 18 and 25 °C) under artificial light and away from heat sources. The monitoring was performed analysing the oils up to 12 months from bottling with quarterly sampling (EV1 at 0 time, EV2 after three months of storage, EV3 after six months of storage, EV4 after nine months of storage, and EV5 after 12 months of storage).

1.2. Methods

Several analyses were regularly performed to determine and quantify any possible evolution of parameters related to the olive oil quality.

1.2.1. Free acidity, peroxide index, and spectrophotometric investigation in the ultraviolet. Free fatty acid content (percent oleic acid), peroxide index (PI expressed as milliequivalents active oxygen per kg oil (meq O_2 per kg)), and UV absorption characteristics (K_{232}, K_{270}, ΔK) were determined in duplicate according to the analytical methods described in EEC (1991) and its subsequent modifications.

1.2.2. Pigments, tocopherols, and biophenols. The determination of pigment (chlorophylls and carotenoids), tocopherol, and biophenol contents were performed according to the methods described in a previous work (Lanza et al., 2013a).

1.2.3. Head-space volatile compounds. The volatile compounds were stripped from 50 g oil samples with N_2 (1.2 $dm^3\ min^{-1}$, for 2 h at 37 °C) using 10 mg 1-nonanol added as the internal standard. The samples were adsorbed onto 50 mg activated charcoal and eluted with 1 ml diethyl ether. Quantitative analysis was carried out using gas chromatography, with a Carlo Erba (Milan, Italy) Mega Series 5160 gas chromatograph equipped with a Restek STABIL-WAX silica capillary column (length 60 m; i.d. 0.32 mm; film thickness 0.5 µm), an on-column injection system, a CO_2 cryogenic accessory to hold the oven at 28 °C, and flame ionisation detector set at 230 °C. The oven temperature programme was run at 28 °C for 6 min, increasing at 1.0 °C min^{-1} to 33 °C (no hold), then 1.8 °C min^{-1} to 110 °C (no hold), and then 2.5 °C min^{-1} to 215 °C, where it was held for 10 min. The carrier gas was H_2 at 40 kPa. The injection volume was 0.5 µl. Quantification was performed by peak area integration with a Carlo Erba Mega Series integrator. Concentrations are expressed as mg kg^{-1} 1-nonanol.

1.2.4. Sensory analysis. The evaluation of the monovarietal olive oils was performed under the conditions described in EEC (1991) and its subsequent modifications (Annex XII) by the CREA-OLI Panel recognized by the International Olive Oil Council (IOC) and the Italian Ministry of Agricultural, Food and Forestry Policies (MIPAAF). Each taster of the panel smelled and tasted the oil sample, in order to analyse the olfactory and gustatory perceptions according to the devised profile sheet of the Annex XII. The attributes evaluated were: fusty/muddy sediment, rancid, musty/humid/earthy, winery/vinegary/acid/sour, metallic, fruity (greenly and ripely), bitter, pungent. The profile sheet uses a 10-point intensity scale that ranges from 0 (no perception) to 10 (extreme). All of these analyses were carried out in duplicate for each sample. To elaborate the sensory data, the method for calculating the median and the confidence intervals was used as detailed in Annex XII, taking into account the attributes with a robust coefficient of variation of 20% or less.

1.2.5. Data analysis and statistics. Chemical data were reported as mean values of two replications. Data were subjected to one-way ANOVA and the differences were compared with Fisher's test at 0.05 probability level.

2. Results and discussion

The evolution of the main chemical and physico-chemical characteristics of the oil during the storage period are reported in Table 1.

The acidity shows a very slight increase, probably due to the release of fatty acids from lipids by hydrolysis.

The MEVOO before the storage had a considerable content of natural antioxidants such as α-tocopherol (168 mg kg^{-1}). After 3 months of storage, the α-tocopherol content decreased consistently (118 mg kg^{-1}), remained constant until the 9th month of storage, then decreased reaching the value of 64 mg kg^{-1} at the end of the 12 months of storage.

Table 1. Chemical and physico-chemical characteristics of MEVOO during storage

Parameters	EV1	EV2	EV3	EV4	EV5
Free fatty acidity (g oleic acid/100 g)	0.29 [a]	0.32 [a]	0.33 [a]	0.31 [a]	0.35 [a]
Peroxide value (mEq O_2 kg^{-1})	11.9 [a]	18.6 [b]	17.3 [b]	11.0 [a]	12.8 [a]
UV spectrophotometry					
K_{232}	1.970 [a]	1.934 [a]	1.928 [a]	1.984 [a]	2.145 [b]
K_{270}	0.111 [a]	0.109 [a]	0.108 [a]	0.129 [a]	0.132 [a]
ΔK	–0.001 [a]	0.002 [a]	0.003 [a]	0.004 [a]	0.003 [a]
Carotenoids (VIS spectrophotometry)					
A_{414}	1.043 [a]	1.026 [a]	1.036 [a]	1.026 [a]	1.009 [a]
A_{450}	0.804 [a]	0.803 [a]	0.805 [a]	0.799 [a]	0.770 [a]
A_{477}	0.636 [a]	0.645 [a]	0.644 [a]	0.627 [a]	0.610 [a]
Chlorophyll (mg kg^{-1})	5 [a]	5 [a]	5 [a]	5 [a]	5 [a]
α-Tocopherol (mg kg^{-1})	168 [a]	118 [b]	114 [b]	119 [b]	64 [c]
γ-Tocopherol (mg kg^{-1})	6 [a]	3 [a]	5 [a]	5 [a]	4 [a]

Mean values within the same row followed by common superscript letters do not differ significantly ($P<0.05$)

The peroxide value increased up to a maximum of 19 (reached at 3 months of storage) and then started to decrease. This behaviour can be explained by an initial increase in hydroperoxides, which are flavourless compounds produced during the primary step of oxidation. Subsequently these compounds give rise to substances responsible for off-flavours (secondary oxidation) (FRANKEL, 1980).

K_{232} coefficient remained nearly constant or slightly increased after 9 months of storage but has not exceeded the limit of 2.50 fixed for the "extra" quality (EV5=2.145). The K_{270} coefficient remained unaltered throughout the rest of the storage period.

Also, pigment (chlorophyll and carotenoid) content remained practically constant during all the storage. The dark-green glass protects chlorophylls from oxidative degradation, absorbing part of the radiation at wavelengths corresponding to their absorption, so that reduces the activity of the initiators of the reaction of hydroperoxide formation, and limits the deterioration of the quality of the oil.

Table 2. Evolution of phenolic compounds of MEVOO during storage.

Phenolic compounds	EV1	EV2	EV3	EV4	EV5
3,4-DHPEA (Hydroxytyrosol)	3.73 ^{a}	5.07 ^{b}	5.67 ^{b}	4.84 ^{b}	5.21 ^{b}
p-HPEA (Tyrosol)	5.18 ^{a}	6.56 ^{b}	7.30 ^{b}	6.84 ^{b}	6.86 ^{b}
Vanillic acid	0.52 ^{a}	0.64 ^{a}	0.59 ^{a}	0.59 ^{a}	0.43 ^{a}
Vanillin	0.11 ^{a}	1.36 ^{b}	1.08 ^{b}	1.25 ^{b}	0.47 ^{c}
p-Coumaric acid	0.00 ^{a}	0.49 ^{b}	0.48 ^{b}	0.23 ^{c}	0.23 ^{c}
Hydroxytyrosyl acetate	2.49 ^{a}	2.61 ^{a}	2.85 ^{a}	0.48 ^{b}	0.35 ^{b}
Ferulic acid	0.00 ^{a}	0.32 ^{b}	0.14 ^{a}	2.46 ^{c}	2.04 ^{c}
o-Coumaric acid	0.31 ^{a}	0.26 ^{a}	0.28 ^{a}	0.26 ^{a}	0.24 ^{a}
3,4-DHPEA-EDA ox	0.71 ^{a}	1.49 ^{b}	2.11 ^{c}	2.26 ^{c}	4.61 ^{d}
3,4-DHPEA-EDA	10.39 ^{a}	7.69 ^{b}	8.07 ^{b}	6.60 ^{c}	6.44 ^{c}
Oleuropein	2.76 ^{a}	4.01 ^{b}	1.88 ^{c}	1.66 ^{c}	1.08 ^{d}
3,4-DHPEA-EA (Oleuropein aglycon)	0.54 ^{a}	0.42 ^{a}	0.53 ^{a}	0.34 ^{a}	0.52 ^{a}
Tyrosyl acetate	0.41 ^{a}	1.38 ^{b}	2.22 ^{c}	0.77 ^{d}	0.45 ^{a}
p-HPEA-EDA ox	3.56 ^{a}	3.71 ^{a}	7.35 ^{b}	8.00 ^{b}	11.10 ^{c}
p-HPEA-EDA (Oleocanthal)	24.01 ^{a}	13.64 ^{b}	12.57 ^{b}	6.70 ^{c}	6.93 ^{c}
Lignans	41.15 ^{a}	50.75 ^{b}	55.89 ^{b}	49.38 ^{b}	31.40 ^{c}
Cinnamic acid	0.75 ^{a}	0.74 ^{a}	1.94 ^{b}	0.74 ^{a}	0.99 ^{a}
p-HPEA-EA (Ligstroside aglycon)	0.64 ^{a}	0.59 ^{a}	1.39 ^{b}	1.57 ^{b}	1.78 ^{c}
3,4-DHPEA, -EA, H ox	0.00 ^{a}	0.41 ^{b}	0.88 ^{c}	1.00 ^{c}	5.08 ^{d}
3,4-DHPEA, -EA, H	14.34 ^{a}	12.02 ^{b}	10.87 ^{c}	5.36 ^{d}	5.60 ^{d}
p-HPEA, -EA, H ox	6.30 ^{a}	11.46 ^{b}	13.30 ^{c}	7.85 ^{d}	8.70 ^{d}
Apigenin	0.61 ^{a}	0.71 ^{a}	0.76 ^{a}	0.76 ^{a}	0.57 ^{a}
Methyl-luteolin	0.66 ^{a}	0.22 ^{b}	0.36 ^{b}	4.63 ^{c}	4.99 ^{c}
p-HPEA, -EA, H	3.34 ^{a}	5.73 ^{b}	5.41 ^{b}	2.17 ^{c}	1.38 ^{d}

Results are expressed as mg kg^{-1} of tyrosol
Mean values within the same row followed by common superscript letters do not differ significantly ($P<0.05$)

During storage, the quantity of main active biophenols (secoiridoids and lignans) of virgin olive oil decreases (Table 2). Secoiridoids developing during crushing from the

hydrolysis of oleuropein and ligstroside include an isomer of the oleuropein aglycon (3,4-DHPEA-EA), the ligstroside aglycon (*p*-HPEA-EA) and the dialdehydic form of decarboxymethyl elenolic acid linked to hydroxytyrosol (3,4-DHPEA) and tyrosol (*p*-HPEA), respectively 3,4-DHPEA-EDA and *p*-HPEA-EDA or oleocanthal. The main lignans found in virgin olive oil are (+)-1-acetoxypinoresinol and (+)-1-pinoresinol. Both secoiridoids and lignans affect the quality of the sensory and health properties of virgin olive oil: 3,4-DHPEA-EA, *p*-HPEA-EA, and, with a minor role, 3,4-DHPEA-EDA are responsible of 'bitter' sensation, while *p*-HPEA-EDA (oleocanthal) determines 'pungent' sensation (Esti et al., 2009). During storage in the tanks, the phenolic composition of extra virgin olive oil is modified by the endogenous enzymatic activities contained in the cloudy phase. Oil filtration partially removes the water and enzymes from extra virgin olive oil, and enables to stabilize the phenolic content during its storage. Simple biophenols, such as *p*-HPEA and 3,4-DHPEA, increase probably due to the aglycon degradation (Table 2). Also, oxidised forms of aglycons increase (Table 2): phenols fight oxidation giving electrons to free radicals and oxidising themselves.

The olive oil aromatic profile changes during storage, due to the neo-formation of volatile compounds responsible for common defect referred to as "rancid" (pentanal, heptanal, and nonanal; Table 3). This runs parallel to the increase in saturated aldehyde hexanal content (from 1.86 to 2.96 mg kg^{-1}; Table 3) in the oxidation process, that in this case could be considered a useful marker of oxidation, since it comes only from the secondary oxidation of the linoleic hydroperoxide radical (Solinas et al., 1987). The composition of the volatile fraction shows, in addition to a slight increase of substances related to rancid defect, a slight decrease in compounds related to fruitiness (*cis*-3-hexenal, 1-hexanol and *cis*-3-hexen-1-ol; Table 3).

Table 3. Evolution of volatile compounds of MEVOO during storage.

Volatile compounds	EV1	EV2	EV3	EV4	EV5
Saturated aldehydes					
2-methyl butanal	0.12 [a]	0.22 [a]	0.19 [a]	0.23 [a]	0.18 [a]
3-methyl butanal	0.32 [a]	0.41 [a]	0.05 [b]	0.05 [b]	0.06 [b]
Pentanal	0.00 [a]	0.00 [a]	0.00 [a]	0.00 [a]	0.09 [b]
Hexanal	1.86 [a]	2.69 [b]	2.05 [b]	2.74 [b]	2.96 [b]
Heptanal	0.00 [a]	0.07 [b]	0.06 [b]	0.06 [b]	0.06 [b]
Octanal	0.01 [a]	0.01 [a]	0.03 [a]	0.07 [a]	0.04 [a]
Nonanal	0.00 [a]	0.00 [a]	0.03 [a]	0.02 [a]	0.13 [b]
Unsaturated aldehydes					
trans-2-pentenal	0.01 [a]	0.03 [a]	0.00 [a]	0.00 [a]	0.00 [a]
cis-3-hexenal	0.16 [a]	0.15 [a]	0.11 [b]	0.11 [b]	0.07 [c]
trans-2-hexenal	30.12 [a]	36.30 [a]	33.57 [a]	33.92 [a]	31.92 [a]
trans-2-heptenal	0.03 [a]	0.03 [a]	0.02 [a]	0.03 [a]	0.02 [a]
trans-2-octenal	0.00 [a]	0.00 [a]	0.00 [a]	0.00 [a]	0.01 [a]
Saturated alcohols					
Ethanol	13.66 [a]	17.34 [b]	13.04 [a]	17.23 [b]	12.88 [a]
1-propanol	0.13 [a]	0.08 [a]	0.07 [a]	0.10 [a]	0.04 [b]

Table 3 continued

Volatile compounds	EV1	EV2	EV3	EV4	EV5
Iso-propanol	0.04 [a]	0.05 [a]	0.20 [b]	0.12 [b]	0.24 [b]
2-Butanol	0.01 [a]	0.03 [a]	0.03 [a]	0.02 [a]	0.03 [a]
Iso-butanol	0.28 [a]	0.34 [a]	0.28 [a]	0.31 [a]	0.27 [a]
2-methyl-3-butanol	0.00 [a]	0.00 [a]	0.00 [a]	0.00 [a]	0.01 [a]
3-methyl-1-butanol	0.93 [a]	1.04 [a]	0.86 [a]	0.94 [a]	0.83 [a]
3-pentanol	0.07 [a]	0.09 [a]	0.02 [a]	0.01 [a]	0.02 [a]
1-pentanol	0.14 [a]	0.15 [a]	0.12 [a]	0.13 [a]	0.14 [a]
2-methyl-2-pentanol	0.00 [a]	0.03 [a]	0.09 [b]	0.09 [b]	0.09 [b]
1-hexanol	2.40 [a]	2.36 [a]	2.11 [b]	2.20 [b]	1.98 [b]
1-heptanol	0.02 [a]	0.01 [a]	0.01 [a]	0.01 [a]	0.01 [a]
Unsaturated alcohols					
1-penten-3-ol	1.20 [a]	1.47 [a]	1.29 [a]	1.46 [a]	1.36 [a]
cis-2-penten-1-ol	0.65 [a]	0.65 [a]	0.65 [a]	0.70 [a]	0.67 [a]
trans-2-penten-1-ol	0.88 [a]	0.89 [a]	0.81 [a]	0.85 [a]	0.79 [a]
cis-2-hexen-1-ol	0.02 [a]	0.02 [a]	0.02 [a]	0.02 [a]	0.01 [a]
trans-2-hexen-1-ol	5.62 [a]	5.65 [a]	5.14 [a]	5.38 [a]	4.84 [b]
cis-3-hexen-1-ol	1.62 [a]	1.64 [a]	1.45 [b]	1.54 [b]	1.40 [b]
trans-3-hexen-1-ol	0.07 [a]	0.07 [a]	0.06 [a]	0.07 [a]	0.06 [a]
cis-1-octen-3-ol	0.02 [a]	0.02 [a]	0.02 [a]	0.02 [a]	0.03 [a]
Esters					
Ethyl acetate	3.09 [a]	3.82 [b]	3.15 [a]	4.30 [b]	3.98 [b]
Isobutyl acetate	0.00 [a]	0.00 [a]	0.01 [a]	0.02 [a]	0.01 [a]
Ethyl isobutyrate	0.00 [a]	0.00 [a]	0.13 [b]	0.13 [b]	0.13 [b]
1-methyl butyrate	0.07 [a]	0.08 [a]	0.13 [a]	0.12 [a]	0.08 [a]
2-methyl butyrate	0.00 [a]	0.00 [a]	0.19 [b]	0.18 [b]	0.15 [b]
Hexyl acetate	0.01 [a]	0.00 [a]	0.00 [a]	0.00 [a]	0.00 [a]
cis-3-hexenyl acetate	0.11 [a]	0.13 [a]	0.10 [a]	0.11 [a]	0.11 [a]
Ethyl pentanoate	0.05 [a]	0.05 [a]	0.04 [a]	0.05 [a]	0.05 [a]
Saturated ketones					
2-butanone	0.18 [a]	0.27 [b]	0.41 [c]	0.43 [c]	0.37 [c]
2-pentanone	1.81 [a]	2.32 [b]	2.02 [a]	2.35 [b]	2.66 [c]
3-hexanone	0.12 [a]	0.15 [a]	0.13 [a]	0.11 [a]	0.13 [a]
2-heptanone	0.08 [a]	0.02 [b]	0.03 [b]	0.04 [b]	0.02 [b]
2-octanone	0.41 [a]	0.41 [a]	0.36 [a]	0.39 [a]	0.36 [a]
Unsaturated ketones					
2-ethylfuran	0.17 [a]	0.23 [a]	0.04 [b]	0.03 [b]	0.07 [b]
2-pentylfuran	0.00 [a]	0.00 [a]	0.02 [a]	0.01 [a]	0.03 [a]
1-penten-3-one	0.40 [a]	0.56 [b]	0.46 [a]	0.48 [a]	0.40 [a]
6-methyl-5-hepten-2-one	0.02 [a]	0.02 [a]	0.02 [a]	0.02 [a]	0.03 [a]

Table 3 continued

Volatile compounds	EV1	EV2	EV3	EV4	EV5
Aliphatic hydrocarbons					
n-octane	1.01 [a]	1.28 [b]	1.14 [b]	1.30 [b]	1.25 [b]
n-nonane	0.03 [a]	0.05 [a]	0.19 [b]	0.41 [c]	0.24 [b]
1-octene	0.13 [a]	0.16 [a]	0.04 [b]	0.07 [b]	0.04 [b]
2-octene	0.00 [a]	0.03 [a]	0.08 [b]	0.09 [b]	0.10 [b]
3-octene	0.02 [a]	0.08 [b]	0.01 [a]	0.00 [a]	0.01 [a]
Pentene dimers	2.71 [a]	2.88 [a]	2.39 [b]	2.40 [b]	2.23 [b]
α-pinene	0.80 [a]	0.86 [a]	0.79 [a]	0.84 [a]	0.77 [a]
Aromatic hydrocarbons					
p-xylene	0.09 [a]	0.13 [b]	0.15 [b]	0.15 [b]	0.17 [b]
Carboxylic acids					
Acetic acid	0.01 [a]	0.19 [b]	0.34 [c]	0.20 [b]	0.12 [b]
Propionic acid	0.03 [a]	0.02 [a]	0.02 [a]	0.02 [a]	0.02 [a]

Results are expressed as mg kg^{-1} of 1-nonanol (internal standard).
Mean values within the same row followed by common superscript letters do not differ significantly ($P<0.05$)

The organoleptic analysis (Table 4) of MEVOO showed the appearance of the rancid defect after 12 months of storage (median of defect=1.5), while fruity, bitter, and pungent properties decreased progressively during storage.

Table 4. Organoleptic analysis of MEVOO during storage

Samplings	Fruity		Bitter		Pungent		Defect (rancid)	
	Median	CV%	Median	CV%	Median	CV%	Median	CV%
EV1	3.0	11.9	2.0	16.8	3.7	14.1	0.0	0.0
EV2	3.1	14.0	2.9	9.0	3.6	8.9	0.0	0.0
EV3	2.8	14.3	2.1	10.4	2.9	11.0	0.0	0.0
EV4	2.6	4.8	1.9	9.8	2.0	13.9	0.0	0,0
EV5	2.3	13.4	1.5	7.1	2.2	6.4	1.5	9.8

3. Conclusions

In conclusion, after one year of storage, an organoleptic alteration reducing fruity, pungent, and bitter attributes, and occurrence of the rancid defect were observed. This alteration is accompanied by a decrease in phenolic substances and tocopherols and an increase in primary and secondary oxidation products. The composition of the volatile fraction showed a slight increase of substances related to rancid defect, a constant trend of compounds related to fruity characteristic, and a slight decrease in alcohols.

The results obtained revealed that the monovarietal extra virgin olive oil studied, packaged in dark-green glass bottles at room temperature (between 18 and 25 °C) under artificial light and away from heat sources, with an intermediate initial content of natural antioxidants, has maintained all of its chemical, physical, and sensory properties within the ranges of "Extra virgin olive oil" class during the first nine months of storage. Only at the last

sampling, to the twelfth month, the product has been downgraded to a "Virgin olive oil" by the sensory evaluation, as a slight defect "rancid" has occurred.

From all of the analyses carried out, it can be concluded that the optimum time of storage of the oil stored as above is up to approximately 9 months.

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