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Original article

Consumers' acceptance and quality stability of olive oil flavoured with essential oils of different oregano species

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Summary The objective of this study was to evaluate chemical and physical stability and consumers' acceptance of olive oil flavoured with oregano essential oils (EOs; Compacto, Cordobes, Criollo and Mendocino). Samples of olive oil were added with 0.05% EO and stored in dark (D) and light (L) conditions for 126 days. Samples with oregano EO had lower lipid oxidation indicator values [K232, K269, peroxide value (PV) and anisidine value], especially in darkness. Olive oil with Cordobes EO in D had the lowest PV (18.71 $meqO_2\ kg^{-1}$). Using prediction equations, 20 $meqO_2\ kg^{-1}$ PV in olive oil should be reached in 34 days in L control sample and in 126 days in the Cordobes EO sample in darkness. Samples with Cordobes and Criollo EOs in darkness had the highest chlorophyll content after 126 days (2.91 and 2.88 mg kg^{-1}, respectively). Sensory analysis showed that oregano EO addition in olive oil was detected by panellists in discriminative test and affected consumer acceptance.

Keywords Antioxidant, consumer, olive, oregano, preservation, stability.

Introduction

In Argentina, oregano is considered the most important aromatic species because of higher crop surface than other aromatic plants. The commercial oregano species grown in Argentina include *Origanum vulgare* spp. *vulgare* (Compacto), *Origanum vulgare* ssp *hirtum* (two clones: Cordobes and Criollo) and *Origanum* × *majoricum* (Mendocino; Torres *et al*., 2010).

Virgin olive oil is highly appreciated for its taste and aroma, as well as for its nutritional properties. Olive oil has a considerable amount of natural antioxidants such as tocopherols, carotenoids, sterols and phenolic compounds, which are important in the prevention of many diseases. The health benefit also comes from its content of vitamins, especially vitamin E, which has been reported to be one of the most effective natural antioxidants (Ravindra, 2000).

Virgin olive oil should be consumed fresh as it is obtained from olives, but it is normally stored in large containers, making it susceptible to lipid oxidation. Free radicals coming from the oxidation of fat and oils are responsible for developing rancid odours and flavours and decreasing the nutritional quality (Owen *et al*., 2000). Consumer perception is an important aspect defining food product quality; in particular, olive oil is a product highly appreciated by its sensory properties. Rancid flavour resulting from lipid oxidation reactions makes food unacceptable to consumers (Nepote *et al*., 2009). The addition of antioxidants is one of the technically simplest ways of reducing fat oxidation, preserving flavour and colour and decreasing vitamin destruction (Karpinska *et al*., 2001).

Oregano essential oils (EOs) have shown functional properties as antioxidant activity (Loizzo *et al*., 2009). Particularly, oregano EO has shown a strong antioxidant activity in some food products such as roasted peanuts and olive oil acting as a natural antioxidant (Asensio *et al*., 2011, 2012; Quiroga *et al*., 2011; Olmedo *et al*., 2012).

The chemical composition of Argentinean oregano species is different, and their inclusion in olive oil helps this product to preserve its sensory properties; but the antioxidant effect of the EOs of different Argentinean oregano species added to olive oil and how the olive oil sensory properties are affected have not been studied yet. The objective of this research was to evaluate, during storage, the chemical and physical properties and consumers' acceptance of olive oil flavoured with

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EOs obtained from different oregano species (Compacto, Cordobes, Criollo, and Mendocino) grown in Cordoba province (Argentina).

Materials and methods

Leaves and flowers of *Origanum vulgare* spp. *vulgare* (Compacto), *Origanum vulgare* ssp *hirtum* (clone Cordobes), *Origanum vulgare* ssp *hirtum* (clone Criollo) and *Origanum* × *majoricum* (Mendocino) were provided for the Facultad Ciencias Agropecuarias, Universidad Nacional de Cordoba, Cordoba, Argentina. Plants were farmed in Capilla de los Remedios (Cordoba, Argentina) and harvested in April (2010). The botanical identification of the tested oregano species was conducted by the Botanical Museum (CORD), Multidisciplinary Institute of Plant Biology, National University of Cordoba, using the identification keys proposed by Iestwaart (1980), Xifreda (1983) and Rouquaud & Videla (2000). Voucher specimens were deposited at the herbarium of Botanical Museum (CORD). Extra virgin olive oil was provided by Finca di Fieno (Camino Las Rosas, km 3, Cruz del Eje, Cordoba, Argentina).

Essential oil extraction

Samples of leaves and flowers were hydrodistilled for 2 h in a Clevenger-type apparatus with a separated extraction chamber. The oregano EOs were kept in dark flask at −18 °C in freezer.

GC-MS analysis

A gas chromatograph, Perkin-Elmer® Clarus 600 (Shelton, CT, USA), coupled with an ion trap mass detector equipped with a capillary column DB-5 (30 m long, 0.25 mm i.d. and 0.25 mm coating thickness) was used for the separation of the components. Ionisation was performed by electron impact at 70 eV. Mass spectral data were acquired in the scan mode in the m/z range of 35–450. The analysis, identification and quantification of the different peaks of oregano EOs were performed according to Asensio *et al.* (2011).

Radical-scavenging activity

Antioxidant activity was measured on the basis of scavenging the stable free radical, 2,2-diphenyl-1-picrylhydrazyl (DPPH), according to Quiroga *et al.* (2011). Tested EOs were added in 3.9 mL of 0.05 mM DPPH methanolic solution, and their final concentrations were 5.77, 2.77, 1.39 and 0.69 μg mL^{-1}. The absorbance of the samples was measured at 517 nm after 30 min on a UV-V Diode Array spectrophotometer (Hewlett Packard TM HP 8452 A, Palo Alto, CA, USA). Inhibition percentage of DPPH radical was calculated according to the following formula:

$$\%\text{DPPH inhibition} = (1 - (A - \text{Ab}/\text{Ao})) \times 100$$

where A is the absorbance of DPPH solution with the EOs, Ab is the absorbance of 60% methanol with the EO, and Ao is the absorbance of DPPH solution. The inhibitory concentration 50% (IC_{50}) was calculated from the curve obtained by plotting the percentage of inhibition vs. the final EO concentrations (Loizzo *et al.*, 2009).

Storage

Olive oils were added with 0.05% (w/w) of EOs of oregano Compacto (Com), Cordobes (Cor), Criollo (Crio) and Mendocino (Men). Two different conditions were maintained: light exposure (L) and darkness (D). Olive oil without oregano EO was prepared as a control sample (C) and stored simultaneously with the treatment samples in both conditions (light exposure and darkness). Light exposure and darkness samples (treatments and control) were stored at room temperature (23 ± 1 °C) for 126 days and removed from storage every 21 days for analysis. The chemical analyses were performed in the same day that the samples were removed from storage. The treatments were:

1. Light exposure treatments (L): olive oil without EO (L-C) and olive oil added with oregano EOs (L-Com, L-Cor, L-Crio and L-Men).
2. Darkness treatments (D): olive oil without EO (D-C) and olive oil added with oregano EOs (D-Com, D-Cor, D-Crio and D-Men).

Chemical analysis of stored samples

Peroxide value (PV), p-anisidine value, specific extinction values (K232 and K268) and free fatty acids (FFAs) were evaluated on samples removed from storage. Peroxide value was evaluated following the AOAC method (AOAC, 2007). Free fatty acid was determined by the titration method according to AOAC (2007) expressing the results as percentage of oleic acid. Anisidine value (AV) was evaluated following the IUPAC method (IUPAC, 1987).

Specific extinction values (K232 and K268) were measured at 232 nm and 268 nm, respectively, using a spectrophotometer UV-V Diode Array, Spectrophotometer Hewlett Packard™ HP 8452 A. The results were reported as the sample extinction coefficient E (1%, 1 cm; COI, 2001).

Chlorophyll and carotenoid contents were measured following the procedures described by Mosquera *et al.* (1991). The chlorophyll and carotenoid fractions were measured in a spectrophotometer at 670 and 470 nm,

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respectively. The concentration of pigments was expressed using the following equations:

$$\text{Chlorophylls}(\text{mg kg}^{-1}) = (\text{Abs}_{670} \times 10^6)/(613 \times 100 \times \text{density}) \quad (1)$$

$$\text{Carotenoids}(\text{mg kg}^{-1}) = (\text{Abs}_{470} \times 10^6)/(2000 \times 100 \times \text{density}) \quad (2)$$

Sensory analysis

Consumer acceptance and discriminative tests were performed on fresh olive oil samples (day 0 of storage). For the consumer test, the panellists (n = 100) were recruited from Cordoba (Argentina) according to the following criteria: (i) people aged between 18 and 65 years, (ii) nonsmokers, (iii) people without food allergies and (iv) people who consumed olive oil at least twice a week. For sample evaluation, 2 mL olive oil samples were placed on a slice (3 × 3 × 1 cm long, wide and thickness) of white bread (Pan Liviano, Fargo SA, Cordoba, Argentina). The samples were served in a cardboard dish coded with 3-digit random numbers. Five olive oil samples (C, Com, Cor, Crio and Men) were presented to the panellists in random order during the test day. Panellists were asked to smell and taste the samples for odour and flavour evaluations. The panellists were instructed to consume the whole sample and then to rinse their mouths with water and eat a slice of Granny Smith apple between samples to minimise any residual effect. A 9-point hedonic scale, ranging from 1 (dislike extremely) to 9 (like extremely), was used to evaluate odour and flavour acceptances from the samples (Meilgaard *et al.*, 2006).

For discriminative tests, duo-trio and directional paired comparison tests were performed on olive oil samples according to Meilgaard *et al.* (2006). Twenty panellists participated for the evaluation. They were recruited from the Food Science department (Facultad de Ciencias Agropecuarias, Universidad Nacional de Cordoba, Cordoba, Argentina). The samples were served as it was described for consumer test. For duo-trio evaluation, panellist received three samples simultaneously; one sample was marked as 'reference' and had the same formulation as one of the coded samples. The panellist was asked to choose the sample that was similar to the reference. For directional paired comparison test, panellists received two samples: one was the control sample and the other was flavoured olive oil sample (Com, Cor, Crio and Men). Both samples were coded and the panellists were asked to identify which of the two samples had oregano EO.

Statistical analysis

Three replications of the experiment were made. The data were analysed using the InfoStat software, version 2011p (Facultad de Ciencias Agropecuarias, Universidad Nacional de Cordoba). Analysis of variance (ANOVA, α = 0.05) and LSD Fisher's multiple range test were performed to determine significant differences between means. The linear regression equations of the chemical variables from the storage study of olive oil (PV, K232 and K268, AV, FFAs and chlorophyll and carotenoid contents) were obtained. Principal component analysis (PCA; Souza *et al.*, 2011; Cruz *et al.*, 2013) was performed on the correlation matrix of the standardised (normalised) data from chemical indicators. Associations between different treatments and quality parameters of olive oil were explored by PCA. Cluster analysis (CA) was carried out to obtain groups of olive oil treatments with similar characteristics (Granato *et al.*, 2011). Sample similarities were calculated on the basis of Euclidean distance, and the groups of olive oil treatments with similar characteristics were obtained using the unweighted pair-group method (UPGMA; Asensio *et al.*, 2012). Means, standard deviations, ANOVA and LSD Fisher's multiple range test were also performed between groups obtained from the CA (Nepote *et al.*, 2009). For consumer test data, ANOVA and LSD Fisher's multiple range test were performed (Cruz *et al.*, 2012). For discriminative test data, statistical tables were used for finding significant differences (Meilgaard *et al.*, 2006).

Results and discussion

Oregano essential oil composition

The major components are listed in Table 1. Only those components that presented concentrations higher than 0.1% are reported. The major components were terpinolene (23.59%), terpinen-4-ol (22.099%) and thymol (13.279%) in Compacto EO; carvacrol (24.89%), terpinen-4-ol (20.38%) and p-cymene (14.81%) in Cordobes EO; carvacrol (30.63%), terpinen-4-ol (16.69%) and o-cymol (8.85%) in Criollo EO; and thymol (20.11%), 3-carene (15.97%) and trans-sabinene hydrate (24.05%) in Mendocino EO. The composition of *Origanum vulgare* spp. *vulgare* and spp. *virens* EOs had been found to be rich in acyclic compounds and sesquiterpenoids (Skoula *et al.*, 1999; Esen *et al.*, 2007). Oregano EOs from Argentina have also been found to be rich in sabinyl compounds (Dambolena *et al.*, 2010). *O. mayoricum* is rich in trans-sabinene hydrate hydrate, thymol and γ-terpinene. With respect to the chemical structure,

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Table 1 Essential oil composition of oregano varieties (Compacto, Cordobes, Criollo and Mendocino) analysed by GC-MS

Retention time	Compounds	Essential oil of oregano – relative percentages (%)			
		Compacto	Cordobes	Criollo	Mendocino
5.89	ρ-Xilene	0.00^a	0^a	0^a	0.73^b
6.19	α-Thujene	0.44^b	0^a	0^a	0.31^c
6.38	α-Pinene	0.54^b	0.8^c	3.8^d	0^a
6.78	Camphene	0.15^b	0^a	0^a	0^a
7.39	Sabinene	0.00^a	0.5^b	0.31^b	0.62^c
7.83	Myrcene	0.00^a	0^a	0.57^b	0^a
8.03	Pseudocumol	0.00^a	0^a	0^a	0.5^b
8.26	α-Phellandrene	2.94^b	0^a	0^a	0^a
8.62	α-Terpinene	0.00^a	3.13^b	3.04^b	2.59^a
8.84	p-Cymene	8.82^b	**14.81^c**	0^a	0^a
8.96	o-Cymol	0.00^a	0^a	**8.85^c**	1.75^b
9.06	Limonene	0.64^b	0^a	0^a	0^a
9.1	1.8 Cineole	0.42^b	4.32^c	0^a	0^a
9.24	β-Ocimene	0.00^a	0^a	4.15^b	0^a
9.92	γ-Terpinene	10.09^b	6.73^a	7.65^a	10.58^b
10.87	Terpinolene	**23.57^a**	0.93^a	0.86^a	0.7^a
11.19	Sabinene hydrate trans	0.00^a	0^a	0^a	**24.05^b**
12.44	Pinene hydrate trans	0.00^a	0^a	0.53^a	0^a
13.4	Borneol	0.73^b	3.82^d	3.35^c	0^a
13.78	Terpinen-4-ol	**22.10^c**	**$20.38b^c$**	**16.69^b**	8.54^a
14.2	α-Terpineol	2.21^a	5.15^c	4.67^b	2.41^a
8.81	γ-Terpineol	0.00^a	0^a	0^a	0.21^b
15.34	Trans-pipertol	0.88^b	0^a	0^a	0^a
15.62	Thymol methyl ether	6.52^c	0.6^a	0.41^a	1^b
15.88	β-Pinene	0.00^a	1.45^b	0^a	0^a
16.43	3-Carene	0.07^a	0^a	0^a	**15.97^b**
16.78	Bergamol	0.00^a	0^a	1.24^b	0^a
17.45	Thymol	**13.28^b**	0^a	0.61^a	**20.11^c**
17.78	Carvacrol	0.00^a	**24.89^c**	**30.63^d**	3.88^b
21.65	Caryophyllene	1.06^a	3.88^b	4.41^c	1.41^a
22.1	Farnesene epoxide	0.20^b	0^a	0.21^b	0^a
22.69	γ-Murolene	0.00^a	0.62^b	0^a	0^a
23.51	δ-Germacrene	0.08^a	0^a	0.89^b	0^a
23.35	γ-Gurjunene	0.00^a	0.44^b	0^a	0.79^c
22.41	Naphthalene	1.47^b	0^a	0^a	0^a
21.95	γ-Elemene	0.74^b	0^a	0.99^c	0^a
24.27	β-Bisabolene	0.00^a	0^a	0.83^b	0^a
26.3	Spathulenol	0.88^b	1.84^c	1.87^c	0^a
26.44	Himachalene epoxide	0.20^b	0^a	0^a	0^a
26.5	Caryophyllene oxide	0.00^a	1.32^b	1.41^b	0^a

The same letter in the row means that there are no significant differences at $\alpha = 0.05$ ($n = 3$). Values in bold mean major compounds of each oregano specie analysed by GC-MS.

hydroxyl groups with ortho and para positions provide antioxidant activity to a molecule. Compounds such as thymol, carvacrol and 4-terpineol present in oregano EOs show this characteristic. Cordobes and Criollo EOs have more than 45% of their chemical composition with these three compounds, suggesting high antioxidant activity. The meta position has little or no effect on the antioxidant property of chemical structures. Phenols with ortho substitution increase the stability of the free radical and hence its antioxidant potential (Muchuweti *et al.*, 2007). These compounds could show an antioxidant activity. Moreover, synergistic activity could take place among the phenolic constituents present in EOs. Kulisic *et al.* (2004) reported that the oxygen-containing fraction is more effective as antioxidant than phenolic fraction or its pure constituents thymol and carvacrol, suggesting that the synergy among minor oxygen-containing compounds has a determining role. This fact may determine a difference in antioxidant activity between similar EOs like Cordobes and Criollo EOs. For this reason, the relationship between the content of particular antioxidants and antioxidant activity is difficult to explain on the basis of only a quantitative analysis of the independent components of a particular EO.

DPPH assay

Radical-scavenging activity of oregano EOs was evaluated by means of the DPPH radical assay. The concentrations that led to 50% inhibition (IC_{50}) are given in Table 2. Samples were able to reduce the stable violet DPPH radical to the yellow DPPH-H, reaching 50% reduction with IC_{50} values ranging from 5.814 μL mL^{-1} in Compacto EO to 1.5814 μL mL^{-1} in Cordobes EO. Cordobes, Criollo and Mendocino EOs had lower IC_{50} values. A low IC_{50} value indicates greater antioxidant activity. This potential antioxidant activity could be related to the chemical composition, especially in Cordobes and Criollo EOs, which showed higher amounts of carvacrol.

Table 2 Regression equations and adjusted R^2 of the DPPH assay and IC_{50} value of essential oils obtained from oregano varieties (Compacto, Cordobes, Criollo and Mendocino)

Essential oil samples	β_0†	β_1†‡	R^2	IC_{50}‡
O. Compacto	13.53	21.89^b	0.97	$5.3134^b \pm 0.34$
O. Cordobes	38.75	11.28^a	0.9	$1.5814^a \pm 0.05$
O. Criollo	34.29	21.24^b	0.96	$2.0776^a \pm 0.05$
O. Mendocino	39.47	12.74^a	0.92	$1.5896^a \pm 0.1$

DPPH, 2,2-diphenyl-1-picrylhydrazyl.

†Regression equations: $Y = \beta_0 + \beta_1 X$, where Y = dependent variable (IC); β_0 = a constant that it is equal to the value of Y when the value of $X = 0$; β_1 = coefficients of X; X = independent variable (percentage of inhibition); R^2: adjusted determination coefficient.

‡ANOVA and LSD Fisher's test: The slope (β_1) of each variable and sample followed with the same letters in the column are not significantly different at $\alpha = 0.05$.

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Storage study for olive oil stability evaluation

The results of the chemical analysis including K232, K268, chlorophyll content (mg kg^{-1}), carotenoid content (mg kg^{-1}), free fatty acid content (FFA,%), PV and p-AV during storage are presented in Table 3. These chemical variables exhibited significant differences between olive oil samples ($\alpha = 0.05$).

K232 is related to the formation of hydroperoxides, conjugated dienes, carboxylic compounds and conjugated trienes. K268 depends on secondary oxidation products formed from the initial compounds detected at 232 nm (Ancin & Martínez, 1991). Samples with the addition of oregano EO stored in darkness exhibited a higher protective effect against lipid oxidation in olive oil for specific extinction values at storage day 126. The highest values for K232 and K268 were found in L-C (3.9 and 0.72, respectively). Treatments in darkness were associated with less oxidation compared to samples exposed to light. In K232, D-Com had the lowest values and did not exhibit significant differences throughout storage. This treatment was followed by D-Crio. The treatment using Criollo EO with light exposure also exhibited a good antioxidant activity, showing the lowest value (K232 of 3.41 at storage day 126) with respect to the other treatments kept under the same conditions. Pokorny *et al.* (2011) reported that the antioxidant effect of a determined molecule is related to its participation in the radical formation in the chain reactions. The inhibitory action of an antioxidant component could be explained for its ability to block the radical chain process. Thymol and carvacrol are isomeric molecules that can take part in the chain reactions initiation during the oxidation of triglycerides from a vegetable oil. In addition, carvacrol may participate in one reaction of chain propagation, but thymol may not. Apparently, thymol is more active and effective antioxidant than carvacrol (Pokorny *et al.*, 2011; Quiroga *et al.*, 2013). The results observed in the present study could be attributed to the chemical composition of this EO (Crio EO) that presented high content in phenols with ortho substitution as carvacrol and terpinen-4-ol, both components known for their antioxidant activity (Muchuweti *et al.*, 2007; Dambolena *et al.*, 2010; Quiroga *et al.*, 2011).

Chlorophyll and carotenoid are mainly responsible for the colour of virgin olive oil that varies from yellow-green to greenish-gold. These compounds play an important role in oxidative stability due to their antioxidant nature in the dark and pro-oxidant activity in the light (Criado *et al.*, 2008). Colour is a sensory property that has a strong influence on food acceptability. For that reason, colour contributes decisively to initial consumer perceptions about food quality (Cerretani *et al.*, 2009). In the present study, the chlorophyll and carotenoid contents (mg kg^{-1}) showed considerable differences, not only between the oregano EO treatments but also according to dark and light exposure conditions. After storage day 63, treatments in darkness showed differences with those under artificial light. At storage day 126, a protective effect of oregano EO was not observed for the chlorophyll content in light-exposed treatments. However, oregano EO treatments in darkness exhibited the highest values for this quality parameter. The chlorophyll contents in D-Cor and D-Crio were the highest (2.91 and 2.88 mg kg^{-1}, respectively). Both EOs showed high content in oxygenated terpenes (carvacrol, terpinen-4-ol, cymol). These components in Cor and Crio EOs could be responsible for the antioxidant activity (Dambolena *et al.*, 2010) and for the protective effect of chlorophyll in olive oil flavoured with these oregano EOs. Previous reports have demonstrated the protective effect of rosemary, thyme and lemon EOs in olive oil, but only after 28 and 55 days of storage (Ayadi *et al.*, 2009). The degradation of the chlorophyll and carotenoid pigments present in olive oil is very complex. The main difficulty in understanding the steps of degradation is that these pigments yield different end products (Criado *et al.*, 2008). Carotenoid compounds are more sensitive to temperature than chlorophyll. The presence of oxygen is an important factor in the degradation of carotenoid compounds; even a low concentration of oxygen leads to the loss of this pigment (Ayadi *et al.*, 2009). It has been reported that, in parallel with the presence of oxygen, free radicals might also accelerate the degradation rate of carotenoids (Criado *et al.*, 2008). Regarding the carotenoid content, olive oil treatments in darkness showed better performance according to the results of this storage study. This effect could be attributed to a lower rate of oxidation in the treatments in darkness compared with the treatments exposed to light. In addition, the oxidation of carotenoids depends on the simultaneous oxidation of unsaturated fats (Ayadi *et al.*, 2009). The unsaturated fats in olive oil were probably oxidised by autooxidation reactions, and the oxidation products thereafter oxidised carotenoid compounds.

Lipid hydrolysis generates FFAs by chemical or enzymatic reactions. This phenomenon is of particular interest in water-containing lipid matrices, such as virgin olive oil. Free fatty acids affect the susceptibility to oxidative degradation and also contribute to the reduction in the shelf life in olive oil (Lozano-Sánchez *et al.*, 2010). In this storage study, FFA values were lower than the limits set by EU Regulation 2568/91 for extra virgin olive oil (Table 3). This lipid deterioration indicator did not show significant differences between samples during storage, despite the fact that these values increased during storage.

The PV was the chemical indicator that demonstrated greater changes during storage. As was observed

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Table 3 Means (n = 3) of chemical variables analysed in olive oil samples at storage days 0, 63 and 126

Storage days	Oregano essential oil†									
	L-C	L-Com	L-Cor	L-Crio	L-Men	D-C	D-Com	D-Cor	D-Crio	D-Men
	K232									
0 day	$1.65 \pm 0.09^{a}_{1}$	$1.65 \pm 0.09^{a}_{1}$	$1.65 \pm 0.09^{a}_{1}$	$1.65 \pm 0.09^{a}_{1}$	$1.65 \pm 0.09^{a}_{1}$	$1.65 \pm 0.09^{a}_{1}$	$1.65 \pm 0.09^{a}_{1}$	$1.65 \pm 0.09^{a}_{1}$	$1.65 \pm 0.09^{a}_{1}$	$1.65 \pm 0.09^{a}_{1}$
63rd day	$3.38 \pm 0.02^{a}_{2}$	$3.37 \pm 0.33^{a}_{2}$	$2.75 \pm 0.35^{a}_{2}$	$2.98 \pm 0.17^{a}_{2}$	$3.07 \pm 0.55^{a}_{2}$	$2.37 \pm 0.55^{a}_{2}$	$2.62 \pm 0.29^{a}_{1}$	$2.73 \pm 0.24^{a}_{2}$	$2.81 \pm 0.15^{a}_{2}$	$2.11 \pm 0.44^{a}_{2}$
126th day	$3.9 \pm 0.39^{e}_{3}$	$3.58 \pm 0.12^{d}_{2}$	$3.62 \pm 0.21^{d}_{3}$	$3.41 \pm 0.26^{c}_{3}$	$3.75 \pm 0.24^{d}_{3}$	$3.17 \pm 0.24^{b}_{3}$	$3.03 \pm 0.04^{a}_{1}$	$3.36 \pm 0.19^{b}_{3}$	$3.23 \pm 0.19^{b}_{3}$	$3.49 \pm 0.13^{c}_{3}$
	K268									
0 day	$0.11 \pm 0.03^{a}_{1}$	$0.11 \pm 0.03^{a}_{1}$	$0.11 \pm 0.03^{a}_{1}$	$0.11 \pm 0.03^{a}_{1}$	$0.11 \pm 0.03^{a}_{1}$	$0.11 \pm 0.03^{a}_{1}$	$0.11 \pm 0.03^{a}_{1}$	$0.11 \pm 0.03^{a}_{1}$	$0.11 \pm 0.03^{a}_{1}$	$0.11 \pm 0.03^{a}_{1}$
63rd day	$0.57 \pm 0.16^{c}_{2\ 3}$	$0.31 \pm 0.07^{a}_{2}$	$0.39 \pm 0.04^{ab}_{2}$	$0.45 \pm 0.09^{b}_{2\ 3}$	$0.38 \pm 0.04^{ab}_{2\ 3}$	$0.33 \pm 0.02^{a}_{2\ 3}$	$0.37 \pm 0.08^{ab}_{2}$	$0.4 \pm 0.06^{ab}_{2}$	$0.41 \pm 0.04^{ab}_{2}$	$0.37 \pm 0.02^{ab}_{2}$
126th day	$0.72 \pm 0.07^{d}_{3}$	$0.6 \pm 0.06^{c}_{3}$	$0.55 \pm 0.09^{b}_{3}$	$0.61 \pm 0.01^{c}_{3}$	$0.47 \pm 0.03^{ab}_{3}$	$0.4 \pm 0.06^{a}_{3}$	$0.56 \pm 0.01^{b}_{3}$	$0.53 \pm 0.01^{ab}_{3}$	$0.54 \pm 0.25^{ab}_{3}$	$0.45 \pm 0.08^{ab}_{2}$
	Chlorophyll content (mg kg^{-1})									
0 day	$3.19 \pm 0.15^{ab}_{1}$	$3.42 \pm 0.04^{b}_{1}$	$2.99 \pm 0.04^{a}_{1}$	$2.99 \pm 0.17^{a}_{1}$	$3.18 \pm 0.09^{ab}_{1}$	$3.19 \pm 0.15^{ab}_{1}$	$3.42 \pm 0.04^{b}_{1}$	$3.16 \pm 0.32^{ab}_{1}$	$3.05 \pm 0.13^{a}_{1}$	$3.22 \pm 0.02^{ab}_{1}$
63rd day	$2.02 \pm 0.17^{b}_{2}$	$1.59 \pm 0.27^{a}_{2}$	$1.39 \pm 0.59^{a}_{2}$	$1.41 \pm 0.28^{a}_{2}$	$1.92 \pm 0.07^{ab}_{2}$	$3.08 \pm 0.28^{c}_{1\ 2}$	$3.08 \pm 0.36^{c}_{2}$	$3.13 \pm 0.08^{c}_{1}$	$3 \pm 0.15^{c}_{1}$	$3.14 \pm 0.15^{c}_{1}$
126th day	$1.47 \pm 0.13^{c}_{3}$	$1.04 \pm 0.08^{ab}_{3}$	$0.98 \pm 0.24^{a}_{2}$	$1.09 \pm 0.04^{ab}_{3}$	$1.2 \pm 0.03^{b}_{3}$	$2.92 \pm 0.09^{f}_{3}$	$2.71 \pm 0.01^{d}_{3}$	$2.91 \pm 0.09^{f}_{2}$	$2.88 \pm 0.03^{e}_{1}$	$2.82 \pm 0.11^{e}_{2}$
	Carotenoid content (mg kg^{-1})									
0 day	$1.44 \pm 0.04^{b}_{1}$	$1.4 \pm 0.07^{b}_{1}$	$1.24 \pm 0.06^{a}_{1}$	$1.4 \pm 0.02^{b}_{1}$	$1.5 \pm 0.17^{b}_{1}$	$1.42 \pm 0.04^{b}_{1}$	$1.44 \pm 0.04^{b}_{1}$	$1.37 \pm 0.05^{ab}_{1}$	$1.4 \pm 0.02^{b}_{1}$	$1.5 \pm 0.17^{b}_{1}$
63rd day	$1.07 \pm 0.06^{a}_{2}$	$1 \pm 0.04^{a}_{2}$	$1.06 \pm 0.13^{a}_{2}$	$1.07 \pm 0.18^{a}_{2}$	$1.09 \pm 0.02^{a}_{2}$	$1.4 \pm 0.08^{b}_{1}$	$1.41 \pm 0.14^{b}_{1}$	$1.3 \pm 0.11^{b}_{1}$	$1.37 \pm 0.09^{b}_{2}$	$1.39 \pm 0.7^{b}_{1\ 2}$
126th day	$0.83 \pm 0.12^{a}_{3}$	$0.76 \pm 0.06^{a}_{3}$	$0.84 \pm 0.04^{a}_{3}$	$0.77 \pm 0.05^{a}_{3}$	$0.82 \pm 0.01^{a}_{3}$	$1.37 \pm 0.08^{b}_{2}$	$1.35 \pm 0.12^{b}_{1}$	$1.31 \pm 0.09^{b}_{1}$	$1.3 \pm 0.08^{b}_{2}$	$1.22 \pm 0^{b}_{3}$
	FFA (%)									
0 day	$0.24 \pm 0.01^{a}_{1}$	$0.24 \pm 0.01^{a}_{1}$	$0.24 \pm 0.01^{a}_{1}$	$0.24 \pm 0.01^{a}_{1}$	$0.24 \pm 0.01^{a}_{1}$	$0.24 \pm 0.01^{a}_{1}$	$0.24 \pm 0.01^{a}_{1}$	$0.24 \pm 0.01^{a}_{1}$	$0.24 \pm 0.01^{a}_{1}$	$0.24 \pm 0.01^{a}_{1}$
63rd day	$0.27 \pm 0.02^{a}_{2}$	$0.28 \pm 0.03^{a}_{2}$	$0.29 \pm 0.01^{a}_{2\ 3}$	$0.29 \pm 0.02^{a}_{1\ 2}$	$0.28 \pm 0.03^{a}_{2}$	$0.29 \pm 0.02^{a}_{2}$	$0.28 \pm 0.05^{a}_{1\ 2}$	$0.3 \pm 0.02^{a}_{2}$	$0.27 \pm 0.02^{a}_{1\ 2}$	$0.28 \pm 0.01^{a}_{2}$
126th day	$0.3 \pm 0.01^{a}_{3}$	$0.29 \pm 0.01^{a}_{2}$	$0.3 \pm 0.02^{a}_{3}$	$0.31 \pm 0.03^{a}_{2}$	$0.31 \pm 0.06^{a}_{2}$	$0.28 \pm 0.01^{a}_{2}$	$0.29 \pm 0.04^{a}_{2}$	$0.31 \pm 0.06^{a}_{2}$	$0.29 \pm 0.06^{a}_{2}$	$0.3 \pm 0.001^{a}_{2}$
	Peroxide value (meqO_2 kg^{-1})									
0 day	$7.51 \pm 2.66^{a}_{1}$	$7.51 \pm 0.98^{a}_{1}$	$7.51 \pm 1.87^{a}_{1}$	$7.51 \pm 2.05^{a}_{1}$	$7.51 \pm 2.1^{a}_{1}$	$7.51 \pm 1.13^{a}_{1}$	$7.51 \pm 2.23^{a}_{1}$	$7.51 \pm 1.25^{a}_{1}$	$7.51 \pm 1.44^{a}_{1}$	$7.51 \pm 0.99^{a}_{1}$
63rd day	$21 \pm 2.66^{c}_{2}$	$20.9 \pm 0.98^{d}_{2}$	$29.9 \pm 1.87^{d}_{2}$	$26.8 \pm 2.05^{d}_{2}$	$28 \pm 2.1^{c}_{2}$	$15.5 \pm 1.13^{b}_{2}$	$10.7 \pm 2.23^{a}_{1}$	$11 \pm 1.25^{a}_{1\ 2}$	$14.9 \pm 1.77^{a}_{2}$	$14.2 \pm 0.99^{b}_{2}$
126th day	$54.54 \pm 2.66^{f}_{3}$	$46.93 \pm 0.98^{e}_{3}$	$44.85 \pm 1.87^{de}_{3}$	$37.23 \pm 2.05^{d}_{3}$	$39.19 \pm 2.1^{de}_{3}$	$29.16 \pm 1.13^{c}_{3}$	$27.3 \pm 2.23^{b}_{2}$	$18.71 \pm 1.25^{a}_{3}$	$27.53 \pm 1.44^{b}_{3}$	$26.31 \pm 0.99^{b}_{3}$
	P-anisidine value									
0 day	$5.91 \pm 0^{a}_{1}$	$5.9 \pm 0^{a}_{1}$	$5.9 \pm 0^{a}_{1}$	$5.9 \pm 0^{a}_{1\ 2}$	$5.9 \pm 0^{a}_{1}$	$5.9 \pm 0^{a}_{1}$	$5.9 \pm 0^{a}_{1}$	$5.9 \pm 0^{a}_{1}$	$6.41 \pm 0^{a}_{1}$	$7.67 \pm 0^{b}_{1}$
63rd day	$6.01 \pm 2.54^{a}_{1}$	$7.16 \pm 0.9^{ab}_{1}$	$7.9 \pm 1.03^{b}_{1}$	$8.58 \pm 1.5^{a}_{2}$	$6.75 \pm 2.64^{c}_{1}$	$10.5 \pm 0.18^{d}_{2}$	$11.1 \pm 0^{de}_{2}$	$14.4 \pm 0^{f}_{2}$	$11.7 \pm 0.07^{e}_{2}$	$8.31 \pm 0.05^{ab}_{1}$
126th day	$37.03 \pm 5.63^{c}_{2}$	$35.33 \pm 3.46^{b}_{2}$	$42.49 \pm 3.54^{d}_{2}$	$43.65 \pm 1.51^{d}_{3}$	$41.22 \pm 0.12^{d}_{2}$	$32.58 \pm 0.14^{ab}_{3}$	$34.63 \pm 0.16^{b}_{3}$	$30.79 \pm 0.29^{a}_{3}$	$35.13 \pm 0.19^{b}_{3}$	$33.75 \pm 0.24^{b}_{2}$

FFA, free fatty acid.

†Treatments: olive oil control sample at light exposure (L-C) and darkness (D-C), olive oil added with oregano essential oil at light exposure (L-Com, L-Cor, L-Crio and L-Men) and darkness (D-Com, D-Cor, D-Crio and D-Men).

The same letter (superscript) in the column for every dependent variable means that there are no significant differences between treatments, and the same number (subscript) in the row means that there are no significant differences between evaluated periods (α = 0.05).

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with other chemical variables, the effect of light exposure on the olive oil samples was also remarkably evident with respect to the samples in darkness. Moreover, the presence of the EOs in olive oil showed an antioxidant activity. The highest PV was for L-C (54.54 meqO_2 kg^{-1}) in the light treatments and for D-C (29.16 meqO_2 kg^{-1}) in the darkness treatments. Between the oregano EO treatments with light exposure (L-Com, L-Cor, L-Crio and L-Men), there were no significant differences in PV. For the treatments in darkness, the lowest PV was in D-Cor (18.71 meqO_2 kg^{-1}). Better antioxidant activity of Cordobes EO is in accordance with the results observed in the other chemical indicators. High phenol content in this EO could explain it (Dambolena *et al.*, 2010). The lipid oxidation stability of flavoured olive oils with the addition of dry herbs or plant parts of thyme, rosemary, lavender, basil, lemon zests, white sage, garlic, menthe and geranium was previously studied (Caponio *et al.*, 2003; Ayadi *et al.*, 2009). In those studies, it was observed different deterioration patterns and different oxidative kinetics. Olive oil flavoured with basil shows less thermal stability, followed by samples flavoured with lemon and thyme, and finally, the most stable oil observed by those authors is olive oil flavoured with rosemary.

The p-AV is an indicator of secondary products of lipid oxidation reactions, including alcohols, carboxylic acids, aldehydes and ketones (Ancin & Martínez, 1991); Frankel, 2005). For p-anisidine, D-Cor exhibited the lowest value, which indicates that the EO of this oregano variety has a good preserving effect on olive oil.

Regression analysis

The regression equations for the dependent variables (K232, K268, carotenoid and chlorophyll contents, FFA, PV and AV) for all treatments are shown in Table 4. The R^2 value was higher than 0.50 in all treatments for PV, AV, K268, K232 (except in D-Com and D-Crio), chlorophyll content (except in CD, D-Cor and D-Crio) and carotenoid content (only in treatments exposed to artificial light). The highest R^2 value was 0.961 for PV in L-Com. Significant differences in the slope (β_1) were found between olive oil samples for K232, K268, chlorophyll and carotenoid contents, PV and AV. FFA did not show significant differences between treatments. The slope was lower in olive oil samples stored in darkness with the addition of oregano EO for all dependent variables. D-Cor and D-Crio were the treatments that had the lowest β_1 for K232, K268, chlorophyll and carotenoid contents, PV and AV. These results indicate that oregano EOs Cordobes and Criollo have higher antioxidant activity and are better for preserving the physical and chemical properties of olive oil. This antioxidant activity could be attributed to the chemical composition of Cordobes and Criollo EOs rich in hydrocarbons with hydroxyl groups with ortho and para positions (thymol, carvacrol and 4-terpineol). Kulisic *et al.* (2004) reported that the oxygen-containing compound fraction is the most potent antioxidant, suggesting that the antioxidant activity of oregano EO is due to more polar constituents. Comparing Cordobes and Criollo EOs, the presence of cymene and cymol could determine a difference in the antioxidant activity because of the oxygenated nature of the first mentioned compound that is higher in Cordobes EO.

The induction period for olive oil oxidation is defined as the time necessary to reach a PV of 70 meqO_2 kg^{-1} (Nissiotis & Tasioula-Margari, 2002). According to European Regulation 2568/91, extra virgin oils must present PVs below 20 meqO_2 kg^{-1}, K268 $\leq$ 0.22 and K232 $\leq$ 2.60. Using the regression equation, a PV of 20 meqO_2 kg^{-1} in olive oil should be reached in storage days 34, 31, 42, 44 and 55 for L-C, L-Com, L-Men, L-Cor and L-Crio, respectively, and in storage days 87, 92, 95, 95 and 126 for D-C, D-Com, D-Men, D-Crio and D-Cor, respectively. Oregano EO has shown antioxidant activity in other food products such as fried-salted and coated peanuts by prolonging the shelf life (Olmedo *et al.*, 2009, 2012). In specie of oregano (*Origanum vulgare* spp *vulgare*), important antioxidant activity was detected for EO added to olive oil (Asensio *et al.*, 2011).

Principal component analysis

The biplot obtained from the two principal components (CP) in the PCA is presented in Fig. 1. The two principal components explained 84.8% of the variability in the samples during 126 days of storage. CP 1 represented 62.9% of the variability. The variables K232, PV and K268 were placed on the right side of the CP1 biplot. The olive oil samples that showed higher values for these variables were also placed to the right side of the biplot (L-Crio, L-Men, L-Cor, L-Com and L-C). Chemical indicators of lipid oxidation (K232, K268 and PV) were positively related and they were related to olive oil treatments exposed to light. A strong association of PV with K268, chlorophyll content and carotenoid content was observed. L-C and L-Com were the closest treatments for the variables K232, PV and K268, whereas L-Crio was not closely associated in the light exposure treatments.

The dispersion of the points showed great variability among samples. Olive oil samples with lower values for the variables related to lipid oxidation were placed on the left side (negatives values in CP 1) of the biplot. D-C, D-Com, D-Crio and D-Men were associated with carotenoid and chlorophyll contents in contrast

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Table 4 Regression equation and adjusted R^2 for the dependent variables: K232 and K268, chlorophyll and carotenoid contents, free fatty acids, peroxide value and p-anisidine value in olive oil samples: control sample at light exposure (L-C) and darkness (D-C), olive oil added with oregano essential oil at light exposure (L-Com, L-Cor, L-Crio and L-Men) and darkness (D-Com, D-Cor, D-Crio and D-Men) during the storage study

Regression coefficients	Oregano essential oils									
	L-C	L-Com	L-Cor	L-Crio	L-Men	D-C	D-Com	D-Cor	D-Crio	D-Men
	K232									
β_0	1.74	1.827	1.659	2.003	1.633	1.708	1.686	1.594	1.658	1.455
β_1	0.016[c]	0.016[d]	0.014[b]	0.012[b]	0.017[d]	0.01[a]	0.011[b]	0.015[c]	0.009[a]	0.015[c]
R^2	0.75	0.809	0.815	0.69	0.812	0.56	0.222	0.862	0.338	0.751
	K268									
β0	0.274	0.117	0.202	0.177	0.229	0.138	0.135	0.155	0.162	0.179
β1	0.04[b]	0.004[b]	0.003[a]	0.004[b]	0.002[a]	0.002[a]	0.004[b]	0.003[a]	0.003[c]	0.002[a]
R^2	0.67	0.782	0.784	0.711	0.5	0.797	0.801	0.78	0.637	0.652
	Chlorophyll content (mg kg^{-1})									
β0	3.079	2.992	2.632	2.922	3	3.087	3.408	3.199	3.066	3.279
β1	−0.014[b]	−0.017[a]	−0.015[a]	−0.017[a]	−0.016[a]	−0.002[d]	−0.005[c]	−0.001[d]	−0.001[d]	−0.004[c]
R^2	0.913	0.872	0.735	0.861	0.93	0.131	0.702	0.023	0.131	0.683
	Carotenoid content (mg kg^{-1})									
β0	1.483	1.458	1.378	1.46	1.516	1.455	1.445	1.372	1.439	1.481
β1	−0.006[a]	−0.006[a]	−0.004[b]	−0.005[b]	−0.006[a]	−0.001[c]	−0.001[c]	−0.001[c]	−0.001[c]	−0.002[c]
R^2	0.898	0.911	0.733	0.932	0.878	0.354	0.209	0.171	0.232	0.595
	FFA (%)									
β0	0.247	0.259	0.256	0.257	0.252	0.265	0.258	0.263	0.261	0.259
β1	3.90E-04[a]	2.90E-04[a]	3.90E-04[a]	4.50E-04[a]	4.30E-04[a]	2.20E-04[a]	2.40E-04[a]	3.80E-04[a]	2.40E-04[a]	3.00E-04[a]
R^2	0.515	0.321	0.619	0.212	0.412	0.2	0.147	0.287	0.138	0.391
	Peroxide value (meqO_2 kg^{-1})									
β0	6.627	10.111	6.268	7.046	10.37	5.596	5.093	6.302	5.21	5.875
β1	0.393e	0.315[d]	0.315[d]	0.237[c]	0.228[c]	0.165[b]	0.157[b]	0.094[a]	0.161[b]	0.149[b]
R^2	0.866	0.961	0.935	0.871	0.869	0.899	0.699	0.806	0.814	0.912
	P-anisidine value									
β0	−1.036	−0.658	−1.809	−2.677	−2.16	1.284	1.142	1.949	0.896	0.496
β1	0.259[b]	0.26[b]	0.293[c]	0.339[d]	0.3[c]	0.246[a]	0.262[b]	0.251[a]	0.259[b]	0.262[b]
R^2	0.793	0.822	0.812	0.828	0.793	0.847	0.85	0.855	0.901	0.766

The same letter in the row means that there are no significant differences between treatments ($\alpha = 0.05$).

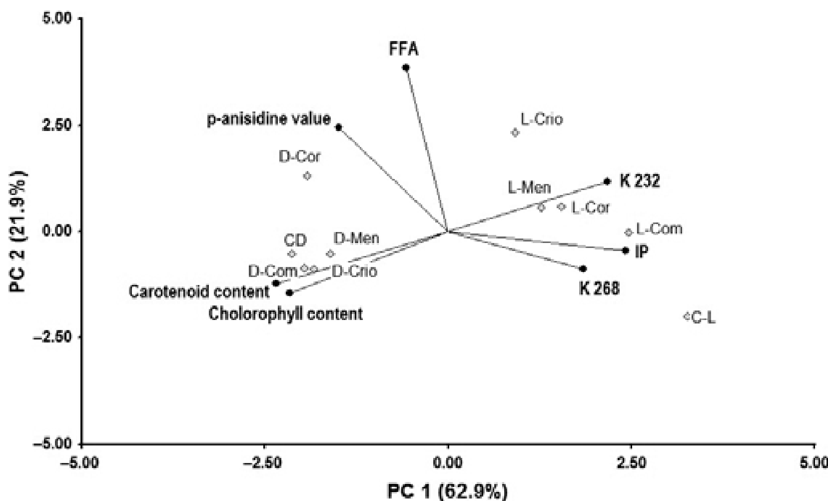

Figure 1 Biplot from the first and second principal components of analysis. Euclidean distance variables: chlorophylls, carotenoids, free fatty acid (FFA), peroxide value (PV), K232, K268 and p-anisidine value (AV). Treatments: olive oil control sample at light exposure (L-C) and darkness (D-C), olive oil added with oregano essential oil at light exposure (L-Com, L-Cor, L-Crio and L-Men) and darkness (D-Com, D-Cor, D-Crio and D-Men).

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with those treatments exposed to light and variables related to lipid oxidation (K232, PV and K268).

Cluster analysis

The CA results of the olive oil samples supplemented with different oregano EOs, considering the dependent variables, are presented as a dendrogram in Fig. 2. Four groups were obtained: Group 1 was formed only by L-C; Group 2 was formed by all treatments in darkness (D-C, D-Com, D-Cor, D-Crio and D-Men); Group 3 was constituted of treatments with the addition of oregano EO exposed to light (L-Cor, L-Men and L-Com), excepting L-Crio, which was placed alone in Group 4. The mean values for the chemical variables by the group of olive oil samples obtained from the CA are presented in Table 5. Significant differences were found between groups for most of the variables. Group 2 (D-C, D-Com, D-Cor, D-Crio and D-Men) showed the lowest K232 and K268 and the highest chlorophyll and carotenoid contents and was significantly different from the other groups. Group 4 (L-Crio) was significantly different from Group 3 considering the abovementioned variables. Therefore, L-Crio showed better protection for olive oil against deterioration in light exposure treatments. In addition, Groups 2 and 4 did not show significant differences, confirming the antioxidant activity of these groups. For AV, there were no significant differences between the treatments.

Consumer and discriminative tests

The results of the consumers' acceptance test are presented in Fig. 3. The flavour and odour acceptance means of the samples were from 5.37 to 6.26 in a 9-point hedonic scale. Significant differences were found between samples. The sample flavoured with Men EO had the highest acceptance value for flavour (6.27 in a 9-point hedonic scale). Com EO was the most accepted sample in odour, but was not significantly different from the other samples except for Cor EO. Olive oil flavoured with Cor EO was the less liked sample for the two analysed attributes in consumer acceptance test, but this sample did not show significant differences in odour and flavour acceptability with respect to control sample.

With respect to discriminative analysis, directional paired comparison and duo-trio tests were performed. In the directional paired comparison test, control sample was compared with samples with oregano EOs (Com, Men, Cor and Crio) with the objective to know whether the addition of oregano EOs is perceived by the panellist in the olive oil. Discriminative panellists detected significantly the samples flavoured with Com, Crio and Cor EOs with 0.1% significance level and the sample flavoured with Men EO with 1% significance level.

The duo-trio discriminative test was performed with the objective to detect differences between samples of olive oil flavoured with the four different species of oregano EOs. The results of this test showed that the panellists were only able to detect differences ($\alpha = 0.05$) when one sample was olive oil flavoured with Men EO in the two samples evaluated. This means that differences in the perception of flavoured olive oil samples were found between Men-Com-, Men-Crio- and Men-Cor-flavoured olive oil samples with a significant level of 5%. The panellists were not able to distinguish any difference between any other olive oil samples flavoured with Com, Cor and Crio EOs.

In previous studies, EO of oregano Compacto demonstrated the antioxidant activity preserving quality parameter in olive oil (Asensio *et al.*, 2011), and it was observed that the addition of EOs preserves the inten-

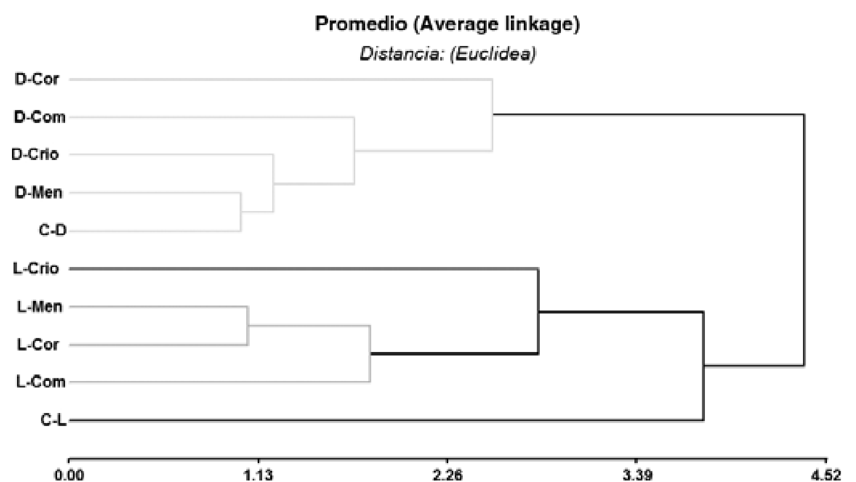

Figure 2 Dendrogram from cluster analysis of olive oil samples (light exposure treatments: L-C, L-Com, L-Cor, L-Crio and L-Men and darkness treatments: D-C, D-Com, D-Cor, D-Crio and D-Men) considering the study variables (chlorophyll and carotenoid contents, free fatty acid, peroxide value, K232, K268 and p-anisidine value).

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Table 5 Means and standard errors from groups of olive oil samples obtained from cluster analysis considering chemical variables

Groups of olive oils treatments	K232			K268			Chlorophyll content (mg kg^{-1})			Carotenoid content (mg kg^{-1})			FFA (%)			Peroxide value (meqO_2 kg^{-1})			p-Anisidine value		
	Means	*n*	E.E.	Means	*n*	E.E.	Means	*n*	E.E.	Means	*n*	E.E.	Means	*n*	E.E.	Means	*n*	E.E.	Means	*n*	E.E.
Group 1	2.77	21	0.16B	0.52	21	0.04B	2.24	21	0.1B	1.12	21	0.04A	0.27	21	0.01A	31.4	21	2.44B	15.31	21	2.8A
Group 2	2.34	84	0.08A	0.33	84	0.02A	3.03	84	0.1C	1.38	84	0.02C	0.28	84	2.90E-03A B	15.2	84	1.26A	17.3	84	1.41A
Group 3	2.7	63	0.09B	0.37	63	0.02A	1.86	63	0.1A	1.12	63	0.02A	0.28	63	3.40E-03A B	27.2	63	1.43B	16.37	63	1.62A
Group 4	2.63	42	0.12B	0.38	42	0.03A	2.51	42	0.1B	1.22	42	0.03B	0.29	42	4.10E-03B	17.1	42	1.73A	18.22	42	1.98A

FFA, free fatty acid. The same letter in the row means that there are no significant differences between treatments (α = 0.05).

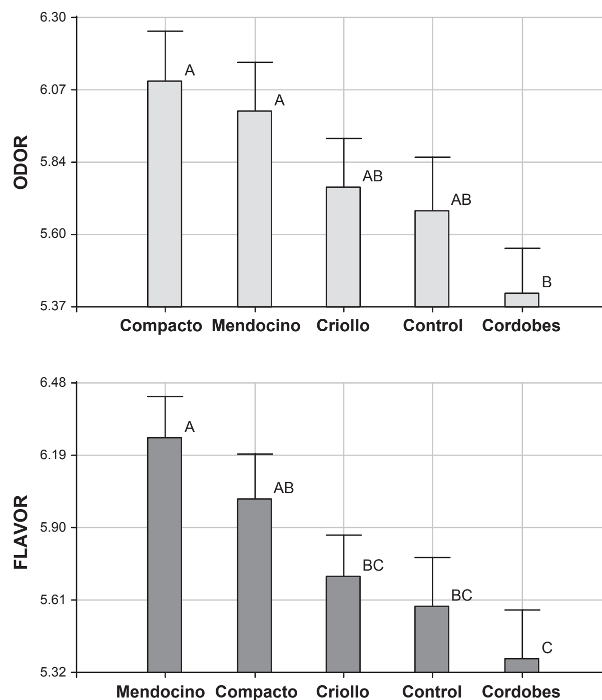

Figure 3 Consumers' acceptance (9-point hedonic scale) of olive oil samples flavoured with oregano essential oils (Compacto, Mendocino, Cordobes and Criollo). The same letter on the bar means that there are no significant differences between treatments (α = 0.05).

sity ratings of positive sensory attribute of olive oil (Asensio *et al.*, 2012). In the present study, EOs of oregano Cordobes and Criollo have higher antioxidant activity preserving chemical parameters related to lipid oxidation in olive oil than the antioxidant activity observed in Mendocino and Compacto EOs. Differences in chemical compositions of oregano EO from Argentine could explain the difference in the antioxidant activity. In addition, it seems that the EOs of oregano that had high activity compounds (Cor and Crio) have lower flavour and odour acceptance by consumers, but this acceptability is similar to the control sample (olive oil without oregano EO addition).

The EOs of four commercial oregano varieties that are cultivated in Argentina have different chemical composition. The oregano EOs of these oregano species have shown a protective effect against deterioration of extra virgin olive oil. Particularly, the oregano EOs of Cordobes and Criollo varieties exhibit a remarkable antioxidant activity. Olive oil with the addition of oregano EO as a quality-preserving agent would not only prevent the product from lipid oxidation extending its shelf life, but also would catch consumer's attention to a nutritional value food. For all these reasons, the food industry should consider the

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use of oregano EO as a potential natural antioxidant for olive oil.

Conclusion

The inclusion of oregano EO provides a protective effect against the deterioration of quality parameters in olive oil. Essential oils of oregano Cordobes and Criollo are a good option in this antioxidant function in olive oil. These EOs of oregano show better preserving effect in olive oil than the other studied oregano EO. Probably, their chemical composition with higher concentration of carvacrol, terpinen-4-ol and p-cymene or o-cymol is responsible for higher antioxidant activity.

Oregano EOs are perceived in the EOs when samples are analysed by discriminative test. However, acceptance test shows that olive oil with the addition of Men and Com EOs has good acceptability, even better than natural olive oil (control sample).

Olive oil is well known and appreciated for its nutritional properties, but is susceptible to lipid oxidation that results in the loss of sensory and nutritional qualities. On the other hand, consumers are demanding and interested in choosing healthier foods. Synthetic antioxidants are thought to be harmful to human health. In this frame, oregano EOs have high potential as preserving agents of olive oil extending the shelf life of this product and also would catch the attention of consumers who look for nutritional value food with functional properties. The food industry might consider the addition of oregano essentials, specially Cordobes and Criollo species, in olive oil as an alternative antioxidant as a preserving agent.

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