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Frying performance of two virgin oils from *Cornicabra* olives with different ripeness indices

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SUMMARY: The frying performance of two virgin olive oils (VOO) from *Cornicabra* olives of different ripeness indices, 2.08 for VOO1 and 4.13 for VOO2, was evaluated. Thermal, oxidative and hydrolytic alterations were determined throughout 40 frying operations with potatoes. The initial oils showed similar fatty acid compositions and oxidative stability indices as determined by Rancimat, but VOO1 presented higher amounts of total polyphenols and tocopherols. The oils showed high and similar frying performance. No significant differences in the levels of polar compounds (PC) were found between the two oils during frying. Therefore, the frying stability *of Cornicabra* VOOs appears to be unconnected with olive fruit ripeness. The limit of degradation at 25% PC as established in different countries was calculated to occur at 55 frying operations in the two oils. As oil toxicity is related to the levels of compounds formed, the use of *Cornicabra* VOOs for frying is highly recommended.

KEYWORDS: Cornicabra *olive fruit; Frying; Oxidative stability; Potatoes; Ripeness index; Virgin olive oil*

RESUMEN: ***Rendimiento en fritura de dos aceites vírgenes de aceitunas* Cornicabra *con diferentes índices de maduración.*** En el presente trabajo se evalúa el comportamiento de fritura de dos aceites de oliva virgen (VOO) obtenidos de aceitunas de la variedad *Cornicabra* con diferentes índices de maduración, 2,08 para VOO1 y 4,13 para VOO2. A lo largo de 40 operaciones de fritura con patatas se determinaron las alteraciones térmicas, oxidativas e hidrolíticas de los aceites. Los aceites iniciales presentaron composiciones de ácidos grasos e índices de estabilidad oxidativa determinados en Rancimat similares entre sí. Sin embargo, las cantidades de fenoles totales y tocoferol fueron más altas para VOO1. Los aceites mostraron una eficacia en fritura elevada y similar. No se encontraron diferencias significativas en los niveles de compuestos polares (PC) durante la fritura entre los dos aceites. Por tanto, la estabilidad en condiciones de fritura de los dos aceites *Cornicabra* no parece estar relacionada con el estado de maduración de las aceitunas. El límite de degradación de 25% de PC establecido en diferentes países se calculó por extrapolación de resultados, alcanzándose éste a las 55 operaciones de fritura para los dos aceites. Debido a que la toxicidad está relacionada con los niveles de compuestos formados, el uso en fritura de aceites de oliva virgen de la variedad *Cornicabra* es altamente recomendado.

PALABRAS CLAVE: *Aceite de oliva virgen; Aceitunas* Cornicabra*; Estabilidad oxidativa; Índice de madurez; Fritura; Patatas*

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1. INTRODUCTION

Virgin olive oil (VOO) has high stability against thermoxidation due to its high content in monounsaturated fatty acids and low levels of unsaturated fatty acids, along with the presence of minor antioxidant components such as phenolic compounds, tocopherols and sterols (Boskou, 2011). However, its composition is affected by factors such as cultivar, location, ripening, harvest period, processing and storage (Velasco and Dobarganes, 2002; Servili *et al*., 2004). Oils obtained from greener olives normally contain larger amounts of antioxidants than those from more mature olives, such as total polyphenols, including oleouropein, hydroxytyrosol and hydroxytyrosol derivatives, and tocopherols (Škevin *et al*., 2003; Yousfi *et al*., 2006; Conde *et al*., 2008). *Cornicabra* is a Spanish variety which normally produces oils with elevated stability to oxidation (Salvador *et al*., 2001a). This is due to a remarkably low content of linoleic acid and elevated amounts of total polyphenols. The influence of fruit ripening on *Cornicabra* virgin oil quality parameters has already been studied (Salvador *et al*., 2001b). Regarding those parameters related to oxidative stability, results showed that oleic acid diminished during ripeness, whereas linoleic acid and free acidity increased. The levels of natural antioxidants and the oxidative stability index presented a more complex behavior.

During frying a complex series of chemical reactions such as polymerization, oxidation and hydrolysis of triglycerides takes place (Dobarganes and Márquez-Ruiz, 2007). Oils which are very rich in essential fatty acids, i.e. linoleic and linolenic acids, can be adequate when consumed raw, but become very unstable at high temperatures originating potential toxic compounds that are ingested and partially absorbed (Dobarganes and Márquez-Ruiz, 2013). On the other hand, the culinary use of very stable oils containing high contents of saturated fatty acids (SFA) could be inadequate from a nutritional point of view (Olivero-David *et al*., 2011). Polar compounds (PC), triglyceride polymers (TGP), polar fatty acids (PFA) and fatty acid composition are good indicators of the quality of used frying fats and oils (Sánchez-Muniz *et al*., 2008; Dobarganes *et al*., 2000). Most European countries have established a maximum PC level of 25 wt%. In addition, a few European countries have adopted a maximum amount of polymeric compounds of 10-12 wt% for oil discarding (DGF, 2000).

In a previous study on three virgin olive oils obtained from *Picual* olives with different ripeness indices, i.e. low, medium and high, the oil obtained from olives with low index was significantly more stable in the discontinuous frying of potatoes (Olivero-David *et al*., 2014). This did not differ substantially in the fatty acid composition, but presented higher amounts of total polyphenols and tocopherols, which gave rise to a higher oxidative stability as measured by the Rancimat test.

In comparison with other varieties, *Cornicabra* shows late maturation; both the pulp and skin remain green for a longer period (Salvador *et al*., 2001a,b). On the basis of a study on changes in oil quality parameters during four successive crop seasons, the best stage of maturity for *Cornicabra* olives has been suggested to be when the ripeness index is higher than 3.0 and lower than 4.0-4.5 (Salvador *et al*., 2001b). To the best of our knowledge, the influence of olive ripeness on the frying performance of *Cornicabra* virgin oils has not yet been studied. *Cornicabra* and *Picual* are known to be two Spanish olive varieties whose virgin oils normally present elevated stability to oxidative degradation (Salvador *et al*., 2001a). As it seems to be for *Picual* virgin oils, it would be of great industrial interest to get to know whether the oils of *Cornicabra* variety also present better frying performance when they come from olives with low ripeness indices (Olivero-David *et al*., 2014).

The main aim of the present study was to evaluate the frying performance of two *Cornicabra* virgin oils obtained from olives with very different ripeness indices. Thermoxidative and hydrolytic alterations, polar and non-polar fatty acid methyl esters, the contents of total polyphenols and tocopherols and oxidative stability in Rancimat were evaluated during the discontinuous frying of fresh potatoes. In addition, the theoretical number of frying operations at which the oils must be discarded was calculated.

2. MATERIALS AND METHODS

2.1. Olive harvesting, ripeness index and oil elaboration

Olives of *Cornicabra* variety were harvested during the 2012/13 olive season in the Agricultural Experimental Station "La Chimenea", Instituto Madrileño de Investigación y Desarrollo Rural, Agrario y Alimentario (IMIDRA), located in the Autonomous Community of Madrid.

About 35 kg of olives were hand-picked in perfect sanitary conditions from olive trees at two ripeness stages: November 19th (VOO1) and December 31st (VOO2). The olive ripeness index was determined according to the method of Uceda and Frias (1975), based on the evaluation of the olive skin and pulp colors. Seven maturity states of the fruit were used in the evaluation:

0, bright-green skin; 1, green-yellowish skin; 2, green skin with reddish spots; 3, reddish-brown skin; 4, black skin with white flesh; 5, black skin with <50% purple flesh; 6, black skin with ≥50% and >100% purple flesh; and 7, black skin and purple flesh.

Olive oils were extracted within a 24-h period using the Abencor system (MC2 Ingenierías y Sistemas, Sevilla, Spain). The olives were crushed with a hammer mill, the paste was mixed at 26 ± 1 ºC for 30 min and then centrifuged at 3,500 rpm for 1 min. The oil was separated by decantation, filtered and stored at 4 ºC in the dark using amber glass bottles prior to analysis.

2.2. Potato frying

Domestic deep-fat fryers with a 1.1 L stainless steel vessel (SOLAC, Vitoria-Gasteiz, Spain) were used for potato frying. *Spunta* variety potatoes (Valencia, Spain) were fried. The initial surface-to-volume ratio of the oil was 0.20 cm^{-1} (225 cm^2/1100 cm^3). The ratio between the amount of potatoes and the volume of frying oil was kept at 183 g/L.1 L during 40 repeated frying operations by replenishing every five frying operations with unused oil in order to maintain, insofar as possible, a constant oil-to-food ratio. The oils were heated for 10 min till 180 ºC and left for 8 min. The potatoes were cut into 2 mm thick slices using a domestic potato slicer, introduced into the oil at 180 ºC and fried for 6 min. The oil was left to cool to 30-35 ºC between same-day frying operations. The cooling time was approximately 4 h between frying operations. On the basis of a previous study (Bastida and Sánchez-Muniz, 2001), a total of 40 frying operations at the rate of 4 fryings per day were performed with each VOO. The total frying time and the entire operation time per day were 24 min and 13 h, respectively. The whole procedure was performed in duplicate using two fryers for each VOO. Thirty-five mL oil from each of the two fryers were taken after 10, 20, 30 and 40 frying operations and kept frozen at -20 ºC until analysis.

2.3. Fatty acid composition

The analysis of fatty acid composition of the oils was performed by GC after derivatization to fatty acid methyl esters (FAME) with 2M KOH in methanol at room temperature (IUPAC, 1992).

The absolute amount of individual FAME was calculated by multiplying the percentage area of each fatty acid methyl ester by the amount of the unaltered FAME fraction expressed as parts per unit giving equivalent results to those obtained using an internal standard (Olivero-David *et al.*, 2014).

2.4. Analysis of polar compounds (PC)

Total PC in the fresh oils and after being used in frying were determined by adsorption chromatography (Dobarganes *et al.*, 2000). One gram of oil was separated in a silica-packed chromatography column using 150 mL of hexane:diethyl ether (90:10, v/v) and 150 mL of diethyl ether to elute the non-polar and polar fractions, respectively. The amount of the non-polar fraction was determined gravimetrically and that of the polar fraction by weight difference.

2.5. High-performance size-exclusion chromatography (HPSEC) analysis

To obtain further information about changes due to thermoxidation and hydrolysis during frying, an HPSEC analysis of the polar fraction was performed following a slight modification of the IUPAC method (Dobarganes *et al.*, 2000). A solution of the polar fraction in tetrahydrofuran (10-15 mg/mL) was analyzed in a High-Performance Liquid Chromatograph (HPLC) (Agilent 1100 series, Madrid, Spain) equipped with a 20-µL loop, two 300 mm x 7.5 mm i.d. (5 µm particle size), 0.01 and 0.05 µm PL gel columns (Agilent, Madrid, Spain), connected in series, operating at 40 ºC, and a refractive index detector (Agilent Technologies 1260 infinity, Madrid; Spain). HPLC-grade tetrahydrofuran was used as the mobile phase with a flow rate of 1 mL/min. Triglyceride oligomers (TGO), triglyceride dimers (TGD), oxidized triglycerides (OTG), diglycerides (DG), monoglycerides (MG) and free fatty acids (FFA) were quantified in the PC fraction. Hydrolytic compounds (HC) were calculated as the sum of DG, MG and FFA, while thermoxidation compounds (TC) were calculated as the sum of TGO, TGD and OTG. Polymers were calculated as the sum of TGD and TGO.

2.6. Isolation and quantification of the altered and unaltered FAME fractions

Samples of the unused and used frying oils were saponified with 0.5M NaOH in ethanol by applying reflux heating during 10 min. Then methylation was performed according to the AOAC (1995) using 20% BF_3 in methanol with reflux heating during 15 min. After methylation, 1 g of sample was separated by adsorption chromatography on silica gel using 150 mL of hexane/diethyl ether (88:12, v/v) and 150 mL of diethyl ether to elute the non-polar (unaltered) and polar (altered) fractions of FAME, respectively (Márquez-Ruiz *et al.*, 1995). As outlined above for the non-polar and polar oil fractions, the amounts of the unaltered and altered FAME fractions were also determined gravimetrically.

Both FAME fractions were analyzed by HPSEC following the same analysis described above for the oil

polar fraction. Thermal fatty acid dimers (thermal-FAD) and non-oxidized fatty acid monomers (non-oxFAM) were quantified in the non-polar fraction, whereas fatty acid oligomers (FAO), oxidized fatty acid dimers (oxFAD), and oxidized fatty acid methyl esters (oxFAM) were quantified in the polar fraction (Márquez-Ruiz *et al.*, 1995). Aliquots of both fractions (10-15 mg/mL tetrahydrofuran) were analyzed in the same HPSEC device and using the same conditions described above for the oil polar fraction.

2.7. Determination of total polyphenolic compounds

Total phenolic compounds in unused and used frying oils were determined after methanol extraction, subsequent reaction with Folin-Ciocalteu reagent and spectrophotometrically determination at an absorption wavelength of 725 nm (Vázquez-Roncero *et al.*, 1975).

2.8. Determination of tocopherols

The determination of α- and γ-tocopherols present in unused and used frying oils was carried out by high performance liquid chromatography according to the IUPAC method (1992).

2.9. Oxidative stability index (OSI)

The unused and used frying oils were analyzed by the Rancimat test to determine the OSI according to AOCS Official Method Cd 12b-92 (Firestone, 1998). A 743 Rancimat device (Metrohm Ltd, Herisau, Switzerland) was used. The OSI was obtained at 100 ºC with a continuous air flow of 20 L/h and using 2.5 g oil.

2.10. Statistical analyses

Unless it is indicated, all determinations were made in triplicate. The Pearson product-moment correlation test was used to find relationships between parameter data and alteration markers in the oils. Linear regressions were performed to ascertain linear adjustments between the concentration of PC, different TC and HC. The SPSS 19.0 statistical program was employed. Comparisons between linear adjustments for the different compounds in the two VOOs were checked by the ANCOVA test using SAS 9.2 statistical program. Statistical significance was set at $p < 0.05$.

3. RESULTS AND DISCUSSION

3.1. Initial characteristics of the oils

Following conventional Madrid-Area harvesting, *Cornicabra* olives were harvested at 25 and 31 weeks after flowering, presenting ripeness indices of 2.08 and 4.13 for VOO1 and VOO2, respectively. These values were relatively lower compared to those reported for other olive varieties, e.g. *Picual*, harvested at the same season period (Olivero-David *et al.*, 2014).

The oils presented very similar fatty acid compositions (Table 1). As expected, the levels of linoleic and linolenic acids were relatively low. The fatty acid composition covered the expected normal range for *Cornicabra* virgin oils, i.e. high levels of oleic acid and low levels of linoleic and stearic acids (Salvador *et al.*, 2001a; b). This differs from VOOs obtained from other varieties (Olivero-David *et al.*, 2014; Sánchez-Casas *et al.*, 2003). *Cornicabra* is one of the Spanish virgin oils with the lowest amount of linoleic acid (Alba-Mendoza, 1996). Beltrán *et al.* (2005) suggested that the fatty acid composition of VOOs is quantitatively affected by two main factors, the olive variety and the ripeness stage. In the present study, despite the differences in the ripeness indices no substantial differences were found in the fatty acid compositions.

The PC levels were low and of the same order in the two oils, although slightly lower for VOO1 (1.90 g/100 g oil) compared to VOO2 (2.70 g/100 g oil). The values obtained reflected the good quality of both oils, as the PC levels for high-quality fresh oils normally range between 0.4 and 6.4 g/100 g oil (Lumley, 1988). The PC levels of the oils agreed with previous data obtained for extra VOOs (Olivero-David *et al.*, 2014; Romero *et al.*, 1995). Similarly to other high-quality olive oils (Bastida and Sanchez-Muniz, 2001), DGs were the major compounds in the polar fraction, followed by OTG. VOO1 showed a lower amount of OTG (0.39 g/100 g oil) than VOO2 (0.61 g/100 g oil). The oils did not differ substantially in the total HC. As expected,

Table 1. Changes in the fatty acid composition (g/100 g oil) of virgin olive oils obtained from *Cornicabra* olives of different ripeness indices after 40 frying operations of fresh potatoes.

	VOO1	VOO1 (40F)	VOO2	VOO2 (40F)
C16:0	11.1	11.0	11.0	11.1
C16:1	1.1	1.0	1.0	1.1
C18:0	2.5	2.6	2.7	2.7
C18:1	78.4	73.8	78.6	73.6
C18:2	3.1	2.2	3.4	2.4
C18:3	0.7	0.4	0.7	0.5
Others	1.2	1.1	1.2	1.2
SFA	13.6	13.6	13.7	13.8
MUFA	79.5	74.8	79.6	74.7
PUFA	3.8	2.6	4.1	2.9

VOO1, VOO2, virgin olive oils from olives with a ripeness index of 2.08 and 4.13, respectively. Results express average values of two determinations. SFA, saturated fatty acids; MUFA, monounsaturated fatty acid; PUFA, polyunsaturated fatty acids.

TGO and TGD were not detected. These are characteristic of oils that have been subjected to high temperatures like refined oils or used frying oils (Dobarganes *et al.*, 1988).

The levels of total polyphenols were within those reported for *Cornicabra* virgin oils (Salvador *et al.*, 2001a). VOO1 showed a remarkably greater content (273.6 mg/kg oil) than VOO2 (61.2 mg/kg oil). The amount of tocopherol, which was mainly constituted by α-tocopherol and also by γ-tocopherol in trace amounts, was also higher in VOO1 (174 mg/kg oil) than in VOO2 (128 mg/kg oil). Therefore, the total amount of antioxidants, i.e. polyphenols and tocopherols, was higher in VOO1.

Both oils presented high resistance to oxidative degradation, showing OSI values greater than 20 h at 100 ºC. Despite the greater amount of antioxidants found in VOO1 and the fact that no substantial differences were found in the fatty acid compositions between the two oils, the oxidative stability of VOO1 ($OSI_{100ºC}$ 24.4 h) was comparable to that of VOO2 ($OSI_{100ºC}$ 25.4 h). These results reflect the complexity of lipid oxidation, which depends not only on the fatty acid composition and levels of antioxidants, but also on a number of other factors (Velasco and Dobarganes, 2002).

Overall, the two oils studied with different ripeness indices presented similar fatty acid compositions, high quality and high and similar oxidative stability, but differed in the total amount of antioxidants, i.e. total polyphenols and tocopherols.

3.2. Oils changes during repeated frying of potatoes

As expected, decreases in the amount of oleic (5.9% and 6.4%), linoleic (29.0% and 29.4%) and linolenic (42.9% and 28.6%) acids were observed in VOO1 and VOO2 after 40 frying operations, respectively. Oleic acid displayed the major decrease in absolute amounts (4.8-5.0 g/100 g of oil). Due to the great differences in the contents of oleic and linoleic acids in olive oils, significant decreases in oleic acid were not detected until linoleic acid was substantially reduced (Romero *et al.*, 1995; 2000). The changes in oleic, linoleic and linolenic acid concentrations were negatively and linearly correlated ($p < 0.05$; $p < 0.001$; $p < 0.001$, respectively) with the number of frying operations (data not shown), which supports studies reported by Romero *et al.* (1995; 2000).

Figures 1 and 2 show the total PC and the TC and HC amounts, respectively. The PC and TC increased linearly (at least $r^2 = 0.981$; $p < 0.001$) with the number of frying operations, whereas, in agreement with previous studies (Bastida and Sánchez-Muniz, 2001; Romero *et al.*, 2003; Velasco *et al.*, 2005), the HC remained constant.

The levels of total PC indicated that the two oils showed similar frying performance. No substantial differences in the total level of alteration were found between the two oils during frying (Figure 1). These results differ from those reported for *Picual* VOOs, which were subjected to the same experimental frying conditions of the

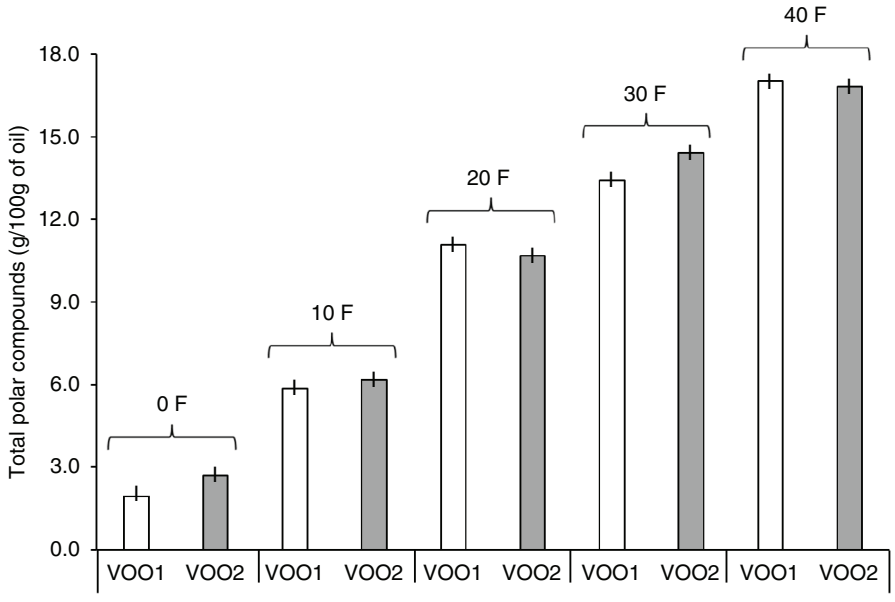

FIGURE 1. Total polar compounds (g/100 g oil) of virgin olive oils (VOOs) obtained from *Cornicabra* olives of two different ripeness indices when unused and after being used in 40 frying operations with potatoes.

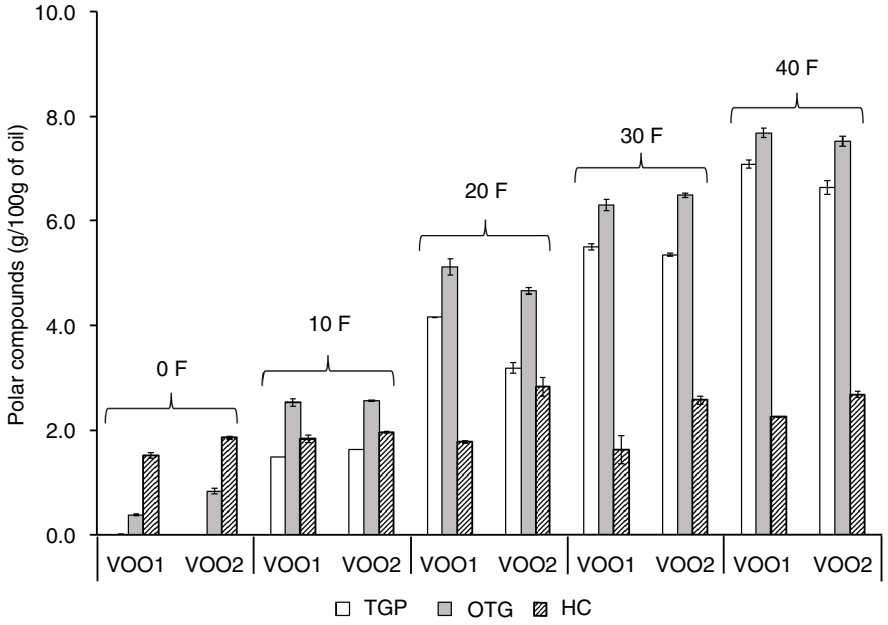

FIGURE 2. Thermoxidative and hydrolytic compounds (g/100 g oil) of the *Cornicabra* virgin olive oils (VOOs). TGP, triglyceride polymers; OTG, oxidized triglycerides; HC, hydrolytic compounds. TGP were calculated as the sum of triglyceride dimers and triglyceride oligomers. HC were calculated as the sum of diglycerides, monoglycerides and free fatty acids.

present study (Olivero-David *et al.*, 2014). The oil of more mature olives exhibited quicker formation of PC, showing values that were greater in 3.5-4 g/100 g oil compared to two oils with lower ripeness index values. While no substantial differences in the fatty acid compositions were found among the three oils, the oil with the lowest ripeness index showed higher antioxidant levels, i.e. total polyphenols and tocopherols (Olivero-David *et al.*, 2014).

The PC and TGP levels found at the end of the frying assay were not greater than 18 g/100 g and 8 g/100 g oil, respectively, which are far from the limits of 25 g/100 g oil for total PC and 10–12 g/100 g oil for TGP (DGF, 2000). This is indicative of the high frying life of *Cornicabra* virgin oils. The values of PC agreed with those of Casal *et al.* (2010) in commercial extra VOO of the same cultivar. This showed the highest stability in the frying of potatoes when compared to a mixture of refined oil and VOO, and refined sunflower oil.

Previous studies suggested that oil thermoxidation changes fit to linear adjustments when frying was performed with low or null oil-turnover but to a power, logarithmic or quadratic adjustment when frequent turnover was done (Sánchez-Muniz *et al.*, 2008). According to the linear total PC level adjustments obtained (Table 2), both VOOs would have been discarded between the 55th and 59th frying operations, respectively. This is relevant taking into account the relatively low turnover of fresh oil performed.

Results of polar FAME are given in Figure 3. From a nutritional point of view, the evaluation of polar and non-polar fatty acyl chains of triglycerides provides valuable additional information about oil degradation (Sánchez-Muniz *et al.*, 2008). Unlike altered triglycerides which comprise both modified and unaltered fatty chains, the analysis of polar and non-polar FAME enables us to know the quantities of the alteration compounds that are directly absorbed after digestion. During frying, the increase in altered fatty acyl chains was linearly adjusted with the number of frying operations (at least $p < 0.01$), whereas the non-polar fraction and the non-oxFAM presented negative linear adjustments (all, $p < 0.01$) (Table 3). Previously, it was suggested that 27.6% PC corresponds to 8.7 to 11.3% polar-ME (Dobarganes and Márquez-Ruiz, 2007). Using 9.5% Polar-ME as a cut-off point, the VOOs shelf-life could have been extended at least to the 64th frying.

TABLE 2. Linear adjustments between different thermoxidation or hydrolytic compounds (g/ 100 g oil) and the number of frying of fresh potatoes with virgin olive oils obtained from *Cornicabra* olives of different ripeness indices.

	Samples[a]	r^2	beta	Intercept[b]	Slope[b]	*p*[c]	VOO1 *vs* VOO2[d] Intercept	VOO1 vs VOO2[d] Slope
PC	VOO1	0.984	0.992	2.269 (1.310, 3.228)	0.381 (0.338, 0.425)	<0.001	NS	NS
	VOO2	0.991	0.995	2.781 (2.479, 3.287)	0.372 (0.340, 0.405)	<0.001		
TGO	VOO1	0.973	0.986	-0.090 (-0.204, -0.023)	0.034 (0.029, 0.039)	<0.001	NS	NS
	VOO2	0.944	0.971	-0.092 (-0.172, -0.025)	0.028 (0.022, 0.034)	<0.001		
TGD	VOO1	0.979	0.989	0.093 (-0.340, 0.526)	0.149 (0.130, 0.169)	<0.001	NS	NS
	VOO2	0.992	0.996	0.058 (-0.188, 0.304)	0.142 (0.131, 0.153)	<0.001		
TGP	VOO1	0.985	0.992	0.003 (-0.451, 0.457)	0.183 (0.163, 0.204)	<0.001	NS	NS
	VOO2	0.994	0.997	-0.034 (-0.300, 0.232)	0.170 (0.158, 0.182)	<0.001		
OTG	VOO1	0.974	0.987	0.689 (0.078, 1.300)	0.188 (0.161, 0.216)	<0.001	NS	NS
	VOO2	0.988	0.994	0.916 (0.535, 1.296)	0.177 (0.160, 0.194)	<0.001		
DG	VOO1	0.438	0.662	1.107 (0.929, 1.285)	0.008 (0.000, 0.016)	0.052	NS	NS
	VOO2	0.637	0.798	1.369 (1.127, 1.611)	0.016 (0.005, 0.027)	0.010		
MG	VOO1	0.358	0.599	0.052 (-0.001, 0.105)	0.002 (0.000, 0.004)	0.089	NS	NS
	VOO2	0.716	0.846	0.043 (0.011, 0.074)	0.003 (0.001, 0.004)	0.004		
FFA	VOO1	0.000	0.008	0.419 (0.321, 0.517)	0.003 (-0.004, 0.000)	0.980	NS	NS
	VOO2	0.229	0.687	0.488 (0.350, 0.627)	0.007 (0.004, 0.013)	0.041		
TC	VOO1	0.981	0.991	0.692 (-0.317, 1.700)	0.371 (0.326, 0.417)	<0.001	NS	NS
	VOO2	0.993	0.997	0.882 (0.320, 1.443)	0.347 (0.322, 0.372)	<0.001		
HC	VOO1	0.309	0.556	1.578 (1.281, 1.874)	0.010 (-0.003, 0.023)	0.120	NS	NS
	VOO2	0.656	0.810	1.900 (1.536, 2.263)	0.025 (0.009, 0.042)	0.008		
TC/HC	VOO1	0.871	0.933	0.595 (-0.885, 2.075)	0.195 (0.128, 0.262)	<0.001	NS	0.020
	VOO2	0.967	0.984	0.622 (0.181, 1.064)	0.122 (0.102, 0.142)	<0.001		

[a]VOO1, VOO2, virgin olive oils from olives with a ripeness index of 2.08 and 4.13, respectively. [b]Values (95 % CI); [c]*p*, linear regression significance; [d]*p*, significant differences between linear adjustments. TGP, triglyceride polymers (oligomers + dimers); TGD, triglyceride dimers; TGO, triglyceride oligomers; OTG, oxidized triglycerides; DG, diglycerides; MG, monoglycerides; FFA, free fatty acids. Hydrolytic compounds (HC), is the sum of DG, MG and FFA. Thermoxidation compounds (TC), is the sum of TGO, TGD and OTG.

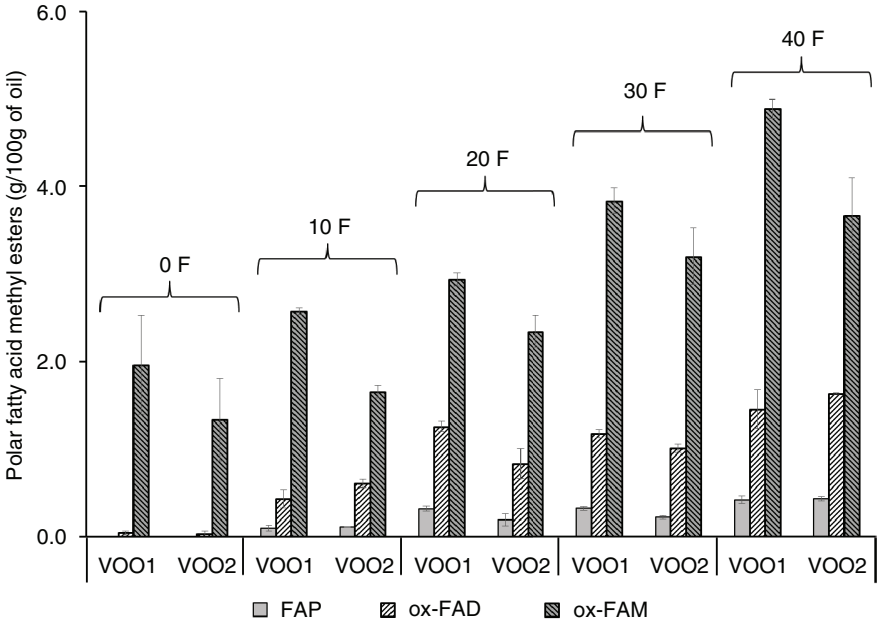

FIGURE 3. Polar fatty acid methyl esters (g/100 g oil) in the oils. FAP, fatty acid polymers; ox-FAD, oxidized fatty acid dimers; ox-FAM, oxidized fatty acid monomers.

TABLE 3. Linear adjustments between polar methyl esters, thermal fatty acid dimers, non-oxidized fatty acid monomers, fatty acid polymers, oxidized fatty acid dimers (g/ 100 g oil) and the number of fryings of fresh potatoes with virgin olive oils obtained from *Cornicabra* olives of different ripeness index.

	Samples[a]	r^2	beta	Intercept[b]	Slope[b]	p[c]	VOO1 *vs* VOO2[d] *Intercept*	VOO1 *vs* VOO2[d] Slope
Polar-ME	VOO1[a]	0.979	0.989	1.994 (1.649, 2.339)	0.117 (0.103, 0.132)	<0.001	0.038	NS
	VOO2	0.966	0.983	1.296 (0.893, 1.699)	0.108 (0.091, 0.124)	<0.001		
Thermal-FAD	VOO1	0.682	0.826	1.135 (-0.183, 2.452)	0.097 (0.043, 0.151)	0.003	NS	NS
	VOO2	0.616	0.785	1.360 (-0.046, 2.766)	0.089 (0.032, 0.146)	0.007		
Non-oxidized FAM	VOO1	0.916	-0.957	96.871 (95.575, 98.168)	-0.214 (-0.267, -0.161)	<0.001	NS	NS
	VOO2	0.893	-0.945	97.344 (95.985, 98.703)	-0.197 (-0.252, -0.141)	<0.001		
FAP	VOO1	0.905	0.951	0.018 (-0.051, 0.087)	0.011 (0.008, 0.013)	<0.001	NS	NS
	VOO2	0.903	0.950	-0.003 (-0.067, 0.061)	0.010 (0.007, 0.012)	<0.001		
OxFAD	VOO1	0.851	0.923	0.161 (-0.136, 0.458)	0.036 (0.023, 0.048)	<0.001	NS	NS
	VOO2	0.939	0.969	0.102 (-0.082, 0.285)	0.036 (0.029, 0.044)	<0.001		
OxFAM	VOO1	0.934	0.967	1.815 (1.439, 2.192)	0.071 (0.056, 0.087)	<0.001	0.049	NS
	VOO2	0.917	0.958	1.198 (0.824, 1.571)	0.062 (0.047, 0.077)	<0.001		
Apolar-ME	VOO1	0.979	-0.989	98.006 (97.661, 98.351)	-0.117 (-0.132, -0.103)	<0.001	0.038	NS
	VOO2	0.966	-0.983	98.704 (98.301, 99.107)	-0.108 (-0.124, -0.091)	<0.001		

[a]VOO1, VOO2, virgin olive oils from olives with a ripeness index of 2.08 and 4.13, respectively. [b]Values (95% CI); [c]*p*, linear regression significance; [d]*p*, significant differences between linear adjustments. Polar ME, Polar methyl esters; Thermal-FAD, Thermal fatty acid dimers; Non-oxidized FAM, Non-oxidized fatty acid monomers; FAP, Fatty acid polymers; OxFAD, Oxidized fatty acid dimers; OxFAM, Oxidized fatty acid monomers; NS, not significant.

Figure 4 shows the levels of total polyphenols and tocopherols. The concentrations of total polyphenols sharply decreased during the first 10 frying operations in the two oils, reaching undetectable levels after the 25th frying operation. The amount of tocopherols also sharply decreased during frying, showing losses of approximately 90% in both oils after the first 5 frying operations. The tocopherol

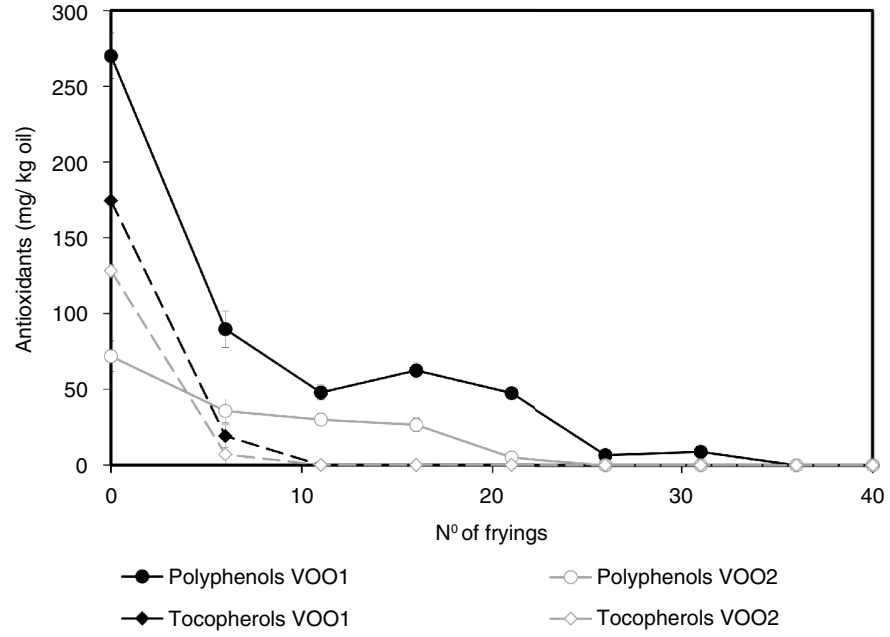

FIGURE 4. Changes in tocopherols and total polyphenols (both in mg/kg oil) of the oils during frying.

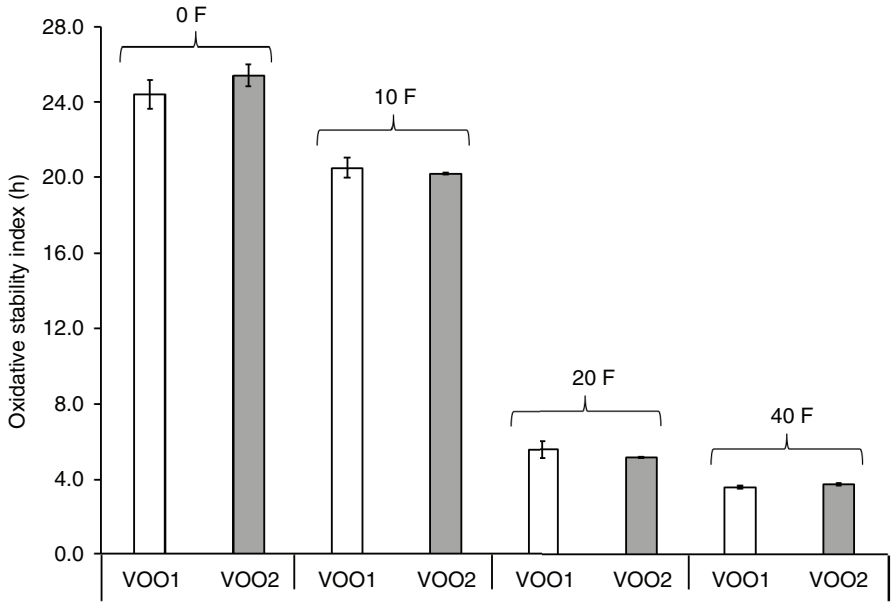

FIGURE 5. Changes in the oxidative stability index (h) measured in Rancimat at 100 ºC after frying.

levels were undetected in the two oils after 10 frying operations.

The results in this study show the rapid degradation of natural antioxidants present in oils at the temperatures normally applied in the frying of foods. In this respect, the similar frying performance of the two oils, mainly differing in the contents of antioxidants, could be related to the rapid degradation of both polyphenols and tocopherols at frying conditions. Barrera-Arellano *et al.* (2002) suggested that the degradation of tocopherols at frying temperatures seems to be predominant over their antioxidative role, as the degradation of tocopherols added to different purified vegetable oils was independent of the degree of unsaturation of the oil. These authors suggested that the protection effect of naturally

occurring tocopherols was a secondary mode of action.

As expected, the OSI values decreased progressively during frying as a consequence of the losses in antioxidants and the accumulation of oxidation compounds (Figure 5). No significant differences were found in the OSI values between the two oils throughout the frying assay. The determination of the oxidative stability of oils by the Rancimat test is a widely used method to determine the resistance to oxidation at the conditions of the test, demanding short analysis time. The oxidative stability index provides information about the oxidative behaviour of oils at room or moderate temperatures if oxygen is not a limiting factor. On the other hand, this index does not allow us to get to know the performance of an oil at the high temperatures applied in the frying of foods because of the different reaction mechanisms involved at frying conditions (Velasco and Dobarganes, 2002). The Rancimat results for the starting oils were similar, even though both oils showed quite different amounts of antioxidants. In contrast, Yousfi *et al*. (2006) found that changes in oil stability and phenolic compounds in olive oils during fruit ripening strongly differed according to the variety and the maturity level of the fruit. The *Cornicabra* virgin oils of the present study showed higher initial oxidative stability than the *Picual* virgin oils tested in a previous study (Olivero-David *et al.*, 2014). This may be attributed in part to the lower amount of linoleic acid and higher content in oleic acid in the *Cornicabra* oils.

4. CONCLUSIONS

Even though the VOO obtained from *Cornicabra* olives with lower ripening index presented higher amounts of polyphenols and tocopherols, the two VOOs studied showed similar frying performance. This seemed to depend more on the low initial level of linoleic acid and the high oleic acid-to-linoleic acid ratio, which were similar for the two oils, than on differences in the polyphenol and/or tocopherol contents. Despite the discontinuous addition of fresh oil, the antioxidants tocopherols and polyphenols rapidly disappeared at frying conditions. As oil potential toxicity is ascribed to the level and composition of the oxidation compounds, the use of *Cornicabra* oils for potato frying is recommended due to their high stability.

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