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Effect of Filtration Systems on the Phenolic Content in Virgin Olive Oil by HPLC-DAD-MSD

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Abstract: This research has been carried out to determine the filtration effect on the phenolic content and on the visual characteristics of olive oil. The influence of the filtration system on the phenolic and water content of virgin olive oil that differed in the year of production, production system and the olive variety was measured. Samples were filtered in the laboratory using two different systems (cotton or filter paper plus sodium sulphate anhydrous). Qualitative and quantitative variation of the phenolic fraction of virgin olive oils was evaluated by HPLC-DAD-MS and correlated with their water content (by Karl Fischer titration) and oxidative stability under forced conditions (by OSI). Colorimetric assays were also carried out in order to calculate the effect of filtration on the visual characteristics of virgin olive oil. After filtration the oxidative stability index decreased and in particular, filtration with cotton showed a significant loss of hydroxytyrosol, a phenol endowed with high antioxidant activity. One interesting behaviour was highlighted: Filtration with either cotton or paper plus anhydrous sodium sulphate led to an apparent increase in the phenolic content. These apparently contradictory data can be explained by considering that the reduction of the water content permits a higher availability of phenolic compounds that remain in oil and are extracted with the methanol-water mixture. Lastly, the filtration of virgin olive oil produced a loss in the intensity of green color and an increase in its lightness.

Key words: Virgin olive oil, phenols, water, filtration, HPLC-DAD-MSD

INTRODUCTION

Polyphenols is a term widely used to designate substances that possess a benzene ring bearing one or more hydroxyl groups, including functional derivatives (Harborne *et al.*, 1989). Polyphenols are polar compounds that can be found in the olive fruit; however many of these compounds are modified or lost during the production process of virgin olive oil (Brenes *et al.*, 1995). The final quantity of polyphenols is also influenced by the cultivar, climatic conditions during growth and degree of ripening (Di Giovacchino *et al.*, 2002; Cerretani *et al.*, 2005). Virgin olive oil is dominated by secoiridoid derivatives, followed by flavonoids and phenolic alcohols. The presence of secoiridoid derivatives provides an indication of the degradation pathways for the phenolic oleosides present in olive paste and wet pomace (Artajo *et al.*, 2007). These derived compounds appear in virgin olive oil and possess antioxidant activity and a lower polarity compared to those in olive fruits (as glycosidic compounds). The partition coefficients between olive oil and water depend on the structure of these compounds and the number of hydroxyl groups.

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The most abundant secoiridoids in virgin olive oil are the dialdehydic forms of elenolic acid linked to hydroxytyrosol or tyrosol (3,4-DHPEA-EDA or *p*-DHPEA-EDA) and an isomer of the oleuropein aglycone (3,4-DHPEA-EA). This can be explained by the fact that DHPEA-EA and 3,4-DHPEA-EDA are the compounds with the highest partition coefficients, as reported by Servili *et al.* (2005).

It is known that olive oil has a low quantity of water (Fregapane *et al.*, 2006; Mendez *et al.*, 2007) and for this reason olive oil can be considered as a water-in-oil emulsion. The presence of phenolic compounds in virgin olive oil and their high antioxidant activity can be explained by the so-called polar paradox (Porter *et al.*, 1989) dictating that: Polar antioxidants are more effective in non polar lipids, whereas non-polar antioxidants are more active in polar lipid emulsions.

Frankel *et al.* (1994) demonstrated that interfacial phenomena are key to a better understanding of antioxidant action in heterogeneous foods and biological systems. The orientation of phenolic compounds in the oil-water interface and the active surface of water droplets influence protection against the oxidation of oil. This study concluded that lipophilic antioxidants are more effective in an oil-in-water emulsion system than in bulk oil, while an opposite trend has been found for hydrophilic antioxidants.

An important factor to consider is the visual appearance of virgin olive oil, as it will strongly influence consumer preference. Color is an intrinsic characteristic of each food product and helps to identify it, to the extent that the consumer is disconcerted if the color changes. From a hedonistic point of view, the color of olive oil can be considered an important organoleptic attribute that is a basic criterion in assessing quality, according to consumer preferences (McEwan, 1994; Pagliarini *et al.*, 1994).

The compounds responsible for the color of virgin olive oil are chlorophylls, carotenoids and flavones (as apigenin and luteolin). Chlorophylls give olive oil its yellow-green color, carotenoids contribute in the yellow-red range (Minguez-Mosquera *et al.*, 1991) and flavones, having an absorbance maximum at around 330-350 nm, provide a yellow color.

Usually, the color of food is measured in L*a*b*. The L*a*b*, or CIE Lab, color space is an international standard for color measurements, adopted by the Commission Internationale d'Eclairage (CIE) in 1976. L* is the luminance or lightness component, which ranges from 0 to 100 and parameters a* (from green to red) and b* (from blue to yellow) are the two chromatic components, which range from -120 to 120 (Papadakis *et al.*, 2000; Segnini *et al.*, 1999; Yam *et al.*, 2004). Nevertheless, the measurement of color is not currently required by regulations established by the European Economic Community (European Union Commission, 1991) to assess the quality of olive oil.

The aim of this report was to evaluate how different filtration processes (normally carried out during virgin olive oil production) affect the characteristics of virgin olive oil. In particular, oxidative stability, water content, the presence of each phenolic compound and color changes of virgin olive oil have been investigated. Eight types of virgin olive oil have been examined that were filtered using two different filtration systems (cotton or filter paper plus anhydrous sodium sulphate).

In our knowledge this is the first study in which olive oils with different origins have been analyzed in order to determine the effects of filtration focusing on the phenolic profile by using a separative technique as HPLC. Furthermore, the filtration systems used have been those that are traditionally applied in small mills.

MATERIALS AND METHODS

Apparatus

All HPLC analyses were performed using a HP 1100 Series instrument (Agilent Technologies, Palo Alto, CA, USA) equipped with a binary pump delivery system, degasser, autosampler, diode-

array UV-VIS Detector (DAD) and Mass-Spectrometer Detector (MSD). The analytical HPLC column used was a C₁₈ Luna column, 5 µm, 25 cm×3.0 mm (Phenomenex, Torrance, CA, USA), with a C₁₈ pre-column (Phenomenex) filter. The mobile phase flow rate was 0.5 mL min⁻¹. All analyses were carried out at room temperature.

The CIELab color space analyses were carried out using a ColorFlex instrument (HunterLab, Reston, VA, USA). To evaluate oxidative stability an eight-channels Oxidative Stability Instrument (OSI) (Omnion, Decatur, IL, USA) was used. The water content of virgin olive oils was obtained using a TitroMatic 1S instrument (Crison Instruments, S.A.; Alella, Barcelona, Spain).

Reagents and Standards

The standard used for HPLC quantification (3,4-dihydroxyphenylacetic acid) was obtained from Sigma-Aldrich Inc. (St. Louis, MO, USA). Methanol, acetic acid, acetonitrile and *n*-hexane were from Merck and Co. Inc. (Darmstadt, Germany). All solvents were HPLC-grade and filtered through a 0.45 µm nylon filter disk (Lida Manufacturing Corp., Kenosha, WI, USA) prior to use. Double-deionized water with a conductivity less than 18.2 MΩ was obtained with a Milli-Q system (Millipore, Bedford, MA, USA). Hydranal-Titran 2 and Hydranal-solvent oil (solvents used to measure the water content with the volumetric titration of Karl Fischer) were from Riedel-deHaën (Seelze, Germany).

Samples

Eight samples of virgin olive oil were obtained from different geographic zones in Italy. They differed in the production year (oxidative state), production plant (traditional and continuous) and storage conditions.

The analysis were carried out on July-September 2006 in the laboratories of the Department of Food Science of the University of Bologna, Cesena (Italy).

Filtration Processes

Two different filtration systems were utilized to reduce the water content of virgin olive oils: (a) cotton and (b) paper and sodium sulphate anhydrous.

- Cotton: Fifty gram of virgin olive oil was passed through 0.5 g of cotton.
- Paper and sodium sulphate anhydrous: Fifty gram of virgin olive oil were passed through filter paper. Next, 100 g of anhydrous sodium sulphate per L of oil was added to the sample and the oil was shaken in order to eliminate the water.

Extraction of Polar Phenolic Fraction

Phenolic compounds were extracted from virgin olive oil by a liquid-liquid extraction method according to Pirisi *et al.* (2000). The dry extracts were dissolved in 0.5 mL of a methanol/water (50/50, v/v) solution and filtered through a 0.2 µm syringe filter (Whatman Inc., Clinton, NJ, USA). The extracts were frozen and stored at -43°C.

Determination of Phenolic Compounds by HPLC-DAD-MSD

Determination of the phenolic fraction was performed using an HPLC-DAD/ESI-MSD equipped with a reverse phase C₁₈ Luna™ column according to Rotondi *et al.* (2004). Phenolic compounds detected at 280 nm were quantified using a 3,4-dihydroxyphenylacetic acid standard calibration curve ($r^2 = 0.9987$). Phenolic compounds were tentatively identified based on their UV-vis and mass spectra (Table 1) obtained by HPLC-DAD/ESI-MSD.

Table 1: Absorption maxima and fragmentation patterns using the ESI interface of the compounds under study

Phenolic compounds	λ_{max} (nm)	Major fragments ESI positive			Other fragments
		m/z [M + 1] ⁺	m/z [M + Na] ⁺	m/z [M + K] ⁺	
Hydroxytyrosol	232/280	-	-	-	137.1
Tyrosol	230/276	-	-	-	121.0
Vanillic acid	228/260/294	169.1	191.1	-	-
Decarboxymethyl-oleuropein aglycon	232/282	-	343.1	359.1	137.1
Pinoretinol	234/280	359.1	381.2	397	398.7
Decarboxymethyl-ligstroside aglycon ⁺					
Acetoxypinoretinol	234/280	-417.1	327.1/439	343.1/455	121.0/385.1
Oleuropein aglycon	232/280	379.1	401.1	417.1	137.1
Ligstroside aglycon	230/276	363.1	385.1	401.1	121.0

Evaluation of Oxidative Stability under Forced Conditions

These analyses were carried out in an eight-channel Oxidative Stability Instrument (OSI) following Carrasco-Pancorbo *et al.* (2005) analytical protocol.

Determination of Water Content in Virgin Olive Oil

The water content was analyzed with a TitroMatic 1S instrument. This measurement uses a Karl-Fischer titration based on a bivalentametric indication (2-electrode potentiometry). A solution of chloroform: Hydranal-solvent oil (a methanolic solvent) 2:1 (v/v) was used to dissolve the sample and Hydranal-Titran 2 was used as a titrating reagent. Each sample was introduced three times and the quantity of sample was measured with the back weighing technique. The sample was dissolved in the solution of chloroform: Hydranal-solvent (2/1, v/v) oil and the titrating reagent was added until the equivalence point. The quantity of water was expressed as mg of water per kg of oil (n = 3).

Statistical Analysis

Data were analyzed using Statistica 6.0 (Statsoft, Tulsa OK, USA) statistical software. The significance of differences at a 5% level between averages was determined by a one-way ANOVA using Tukey's test.

RESULTS AND DISCUSSION

Effect of Filtration on the Water Content

In most olive oil samples the water content decreased significantly after filtration (Table 2). With the exception of sample 7, the control sample always had a higher water content, whereas samples that had been filtered with cotton showed a significant decrease in the quantity of water. However, samples that had been filtered using paper and anhydrous sodium sulphate presented a significant decrease in only four of the eight samples (samples 2, 3, 4 and 8).

Filtration with cotton, termed filtration Bari style, is especially widespread in the olive oil industry plants located in the South of Italy and as is shown from experimental data in Table 2, can be considered effective in reducing the water content.

Evaluation of Oxidative Stability under Forced Conditions

The Oxidative Stability Index (OSI) decreased after the filtration with cotton and paper plus anhydrous sodium sulphate (Table 2). This effect was more pronounced in samples that showed a higher OSI value. For example, the OSI of sample 2 decreased about 4.4% with cotton filtration and by 18.0% with anhydrous sodium sulphate plus paper; sample 6, another control sample with a high oxidative stability, showed a reduction of 15.0% with cotton filtration and 19.0% with anhydrous sodium sulphate plus paper. However samples 4 and 5 that had a low OSI value compared to the

Table 2: Analytic results of virgin olive oils: Oxidative stability (OSI, RSD% = 2.3), Water content (mg of water per kg of olive oil) and Phenolic compounds (mg of analyte as 3,4-dihydroxyphenylacetic acid per kg of olive oil). Quantification of the Individual Components (n = 7) (\pm SD)

		OSI (h)	H ₂ O	HYTY	TY	VA
Sample 1	Control sample	18.35	1070.0 \pm 92.4 (a)	44.7 \pm 1.9 (a,b)	50.1 \pm 1.6 (a)	1.6 \pm 0.1 (b)
	Cotton	16.65	699.9 \pm 34.2 (b)	37.5 \pm 3.4 (b)	57.6 \pm 3.8 (a)	0.7 \pm 0.2 (c)
	Paper+SSA	16.25	935.4 \pm 9.7 (a)	51.5 \pm 4.7 (a)	58.9 \pm 5.5 (a)	5.1 \pm 0.8 (a)
Sample 2	Control sample	27.25	1089.8 \pm 104.8 (a)	8.7 \pm 0.6 (a)	4.2 \pm 0.3 (a)	0.8 \pm 0.4 (b)
	Cotton	26.05	668.9 \pm 12.1 (b)	5.2 \pm 0.2 (a)	3.7 \pm 0.2 (a)	0.7 \pm 0.3 (b)
	Paper+SSA	22.35	806.4 \pm 8.2 (b)	8.5 \pm 0.9 (a)	4.2 \pm 0.4 (a)	1.6 \pm 0.5 (a)
Sample 3	Control sample	14.45	1584.8 \pm 16.3 (a)	13.0 \pm 0.7 (a)	40.0 \pm 2.1 (a)	0.8 \pm 0.2 (a)
	Cotton	13.30	780.1 \pm 49.8 (b)	5.9 \pm 0.3 (c)	23.9 \pm 2.1 (b)	1.0 \pm 0.3 (a)
	Paper+SSA	12.95	702.8 \pm 50.4 (b)	8.7 \pm 0.3 (b)	25.2 \pm 0.7 (b)	1.0 \pm 0.1 (a)
Sample 4	Control sample	8.65	1312.4 \pm 51.4 (a)	1.9 \pm 0.1 (b)	36.0 \pm 1.7 (a)	0.5 \pm 0.2 (c)
	Cotton	8.40	740.1 \pm 39.1 (b)	1.0 \pm 0.0 (c)	25.5 \pm 1.0 (c)	0.8 \pm 0.1 (b)
	Paper+SSA	8.45	796.5 \pm 18.2 (b)	3.8 \pm 0.1 (a)	30.6 \pm 1.1 (b)	1.2 \pm 0.1 (a)
Sample 5	Control sample	7.45	1136.0 \pm 120.3 (a)	2.0 \pm 0.5 (a,b)	5.8 \pm 0.9 (a)	1.8 \pm 1.3 (b)
	Cotton	6.85	786.4 \pm 30.2 (b)	1.5 \pm 0.2 (b)	7.1 \pm 1.3 (a)	2.4 \pm 0.5 (b)
	Paper+SSA	6.35	1033.9 \pm 34.7 (a)	2.4 \pm 0.3 (a)	6.9 \pm 1.1 (a)	5.3 \pm 1.2 (a)
Sample 6	Control sample	20.70	790.0 \pm 46.9 (a,b)	6.9 \pm 0.6 (a)	5.0 \pm 0.7 (a)	3.6 \pm 0.4 (c)
	Cotton	17.60	727.4 \pm 19.6 (b)	5.0 \pm 0.3 (b)	5.7 \pm 0.5 (a)	5.7 \pm 0.1 (b)
	Paper+SSA	16.70	859.4 \pm 17 (a)	7.6 \pm 0.7 (a)	6.2 \pm 0.6 (a)	10.2 \pm 0.9 (a)
Sample 7	Control sample	19.65	975.3 \pm 110.6 (a)	23.3 \pm 0.9 (a)	39.7 \pm 1.3 (a,b)	1.1 \pm 0.1 (b)
	Cotton	19.45	810.1 \pm 17.2 (a)	18.7 \pm 1.1 (b)	41.7 \pm 2.2 (a)	3.5 \pm 0.3 (a)
	Paper+SSA	16.30	861.2 \pm 57.4 (a)	22.2 \pm 1.1 (a)	37.6 \pm 2.4 (b)	3.4 \pm 0.2 (a)
Sample 8	Control sample	17.20	1369.5 \pm 17.6 (a)	3.1 \pm 1.5 (a,b)	7.6 \pm 1.0 (b)	3.5 \pm 1.4 (a)
	Cotton	16.55	768.5 \pm 82.2 (b)	7.9 \pm 1.9 (a)	19.2 \pm 3.0 (a)	nd
	Paper+SSA	14.70	720.6 \pm 29.3 (b)	2.9 \pm 0.9 (b)	7.9 \pm 1.0 (b)	4.7 \pm 1.2 (a)

Table 2: Continued

		DMOA	Pin	DLA+AcPin	Ol Agl	LA
Sample 1	Control sample	12.8 \pm 8.7 (a)	nd	21.7 \pm 3.9 (b)	35.6 \pm 9.6 (a)	10.9 \pm 2.5 (b)
	Cotton	9.1 \pm 2.1 (a)	5.2 \pm 0.7 (a)	26.2 \pm 2.3 (a,b)	37.5 \pm 1.9 (a)	14.9 \pm 1.0 (a)
	Paper+SSA	9.3 \pm 0.7 (a)	4.2 \pm 2.8 (a)	30.5 \pm 2.3 (a)	33.3 \pm 1.4 (a)	14.3 \pm 0.9 (a)
Sample 2	Control sample	18.7 \pm 2.1 (a)	12.0 \pm 2.1 (a)	16.7 \pm 1.5 (c)	22.8 \pm 2.2 (a)	4.9 \pm 1.1 (a,b)
	Cotton	13.5 \pm 1.2 (b)	nd	29.8 \pm 2.8 (a)	15.7 \pm 1.2 (b)	3.5 \pm 0.7 (b)
	Paper+SSA	14.6 \pm 1.7 (b)	12.6 \pm 1.1 (a)	21.7 \pm 2.7 (b)	20.4 \pm 2.4 (b)	5.8 \pm 0.9 (a)
Sample 3	Control sample	3.4 \pm 0.9 (a)	5.8 \pm 0.9 (a)	24.6 \pm 2.5 (a)	10.7 \pm 2.9 (a)	2.8 \pm 0.9 (a)
	Cotton	2.2 \pm 0.5 (a)	2.9 \pm 2.5 (a)	22.2 \pm 8.4 (a)	8.6 \pm 4.4 (a)	3.5 \pm 2.1 (a)
	Paper+SSA	3.1 \pm 0.6 (a)	4.7 \pm 0.3 (a)	26.2 \pm 1.2 (a)	8.8 \pm 1.0 (a)	5.1 \pm 0.9 (a)
Sample 4	Control sample	3.6 \pm 0.4 (a)	4.9 \pm 0.7 (a)	25.8 \pm 3.1 (a)	6.8 \pm 0.6 (a)	4.6 \pm 1.4 (b)
	Cotton	1.5 \pm 0.3 (c)	4.3 \pm 0.4 (a)	28.5 \pm 2.6 (a)	7.1 \pm 0.4 (a)	5.9 \pm 0.7 (a,b)
	Paper+SSA	2.6 \pm 0.4 (b)	4.4 \pm 0.3 (a)	26.6 \pm 0.9 (a)	8.0 \pm 0.8 (a)	7.0 \pm 0.3 (a)
Sample 5	Control sample	4.3 \pm 1.3 (a,b)	4.00 \pm 0.94 (a)	11.4 \pm 2.2 (b)	3.9 \pm 0.8 (b)	0.4 \pm 0.1 (b)
	Cotton	3.4 \pm 0.3 (b)	4.89 \pm 0.77 (a)	18.3 \pm 2.6 (a)	6.6 \pm 1.4 (a)	9.9 \pm 1.8 (a)
	Paper+SSA	5.6 \pm 0.9 (a)	5.70 \pm 0.92 (a)	22.2 \pm 3.8 (a)	6.9 \pm 0.8 (a)	1.5 \pm 0.1 (b)
Sample 6	Control sample	6.3 \pm 2.0 (a)	1.16 \pm 1.35 (b)	27.6 \pm 1.1 (a)	10.2 \pm 0.3 (a)	1.2 \pm 0.0 (b)
	Cotton	7.1 \pm 0.5 (a)	3.11 \pm 0.84 (a,b)	30.9 \pm 4.3 (a)	11.0 \pm 1.6 (a)	2.4 \pm 1.0 (a)
	Paper+SSA	5.6 \pm 0.5 (a)	3.54 \pm 0.27 (a)	32.5 \pm 2.7 (a)	10.4 \pm 0.4 (a)	2.3 \pm 0.2 (a,b)
Sample 7	Control sample	0.9 \pm 1.1 (c)	5.65 \pm 0.29 (a)	7.9 \pm 0.4 (b)	32.1 \pm 0.8 (a)	18.8 \pm 0.3 (a)
	Cotton	2.4 \pm 0.4 (b)	4.61 \pm 0.51 (b)	10.4 \pm 0.9 (a)	28.2 \pm 2.1 (b)	15.7 \pm 1.5 (b)
	Paper+SSA	4.3 \pm 0.3 (a)	4.25 \pm 0.18 (b)	9.8 \pm 0.5 (a)	22.4 \pm 1.2 (c)	12.6 \pm 0.6 (c)
Sample 8	Control sample	16.2 \pm 2.2 (b)	nd	66.4 \pm 9.2 (b)	23.0 \pm 3.6 (b)	17.6 \pm 3.9 (b)
	Cotton	25.4 \pm 4.8 (a)	nd	141.0 \pm 15.3 (a)	36.2 \pm 3.4 (a)	31.0 \pm 4.5 (a)
	Paper+SSA	11.9 \pm 1.2 (b)	nd	59.1 \pm 1.8 (b)	15.8 \pm 1.1 (c)	13.0 \pm 3.0 (b)

Paper+SAA, Filtration by paper and sodium sulphate anhydrous; HYTY, Hydroxytyrosol; TY, tyrosol; VA, Vanillic acid; DMOA, Decarboxymethyl oleuropein aglycon; Pin, pinoselinol; DLA+AcPin, Decarboxymethyl ligstroside aglycon+acetoxypinoselinol; Ol Agl, Oleuropein aglycon; LA, Ligstroside aglycon, Letter(s) a-c in brackets indicate statistically significant differences (HSD Tukey $p < 0.05$)

control sample, demonstrated a decrease of 2.9 and 8.1%, respectively, with cotton filtration and a reduction of 2.3 and 14.8%, respectively, with anhydrous sodium sulphate plus paper filtration.

Considering these results, it can be surmised that the oxidative stability of virgin olive oils is lower when the water content is decreased (after filtration), which is related either to a loss of phenolic compounds or a reduction in their antioxidant activity. The decrease of antioxidant activity, as

mentioned before, can be explained by the polar paradox. In fact, phenolic compounds, being polar molecules, have a higher activity in a water-in-oil emulsion. However after filtration the water content is reduced. As a consequence, the antioxidant capacity of these compounds diminishes, probably due to their particular orientation around small droplets of water.

Evaluation of Behavior of Individual Phenolic Compounds after Filtration

Hydroxytyrosol, decarboxymethyl oleuropein aglycon and oleuropein aglycon are, in that order, the phenolic molecules of virgin olive oil having the highest antioxidant activity (Carrasco-Pancorbo *et al.*, 2005). In general, hydroxytyrosol showed a significant decrease after filtration with cotton with respect to the control sample. This behavior can be explained considering the partition coefficient between olive oil and water of this compound ($K_p = 0.01$ as reported Servili, 2005), which makes it more soluble in water than other phenols.

As reported in Table 2, the concentration of several phenolic compounds seemed to increase after filtration, but is related to the fact that filtration reduces the water content even though the loss of phenolic compounds is not proportional. In fact, it is assumed that the majority of phenolic compounds located around water droplets remain in olive oil.

It is also possible to hypothesize that extraction of phenolic compounds in control samples does not allow for complete recovery of these analytes; indeed, when the analytes are in a more polar matrix the affinity of phenolic extraction to the solvent (methanol/water, 60/40, v/v) is lower and their separation is more difficult. On the other hand, if the extraction with a hydroalcoholic solution is done after the partial elimination of water, phenols are more available to the solvent mixture.

This study represents a novelty in the fact that this apparently increase of phenolic content has been explained by the performance of a study about the variation in water content of virgin olive oil. This type of study has never been considered before by other investigations carried out about filtration effect (Fregapane *et al.*, 2006).

Filtration Effect on the Colorimetric Parameters

A large amount of the particles in suspension was retained by the filtration system. As shown in Table 3, the luminosity of olive oil (L^* value) increased after filtration. Furthermore, when the control

Table 3: Values of $L^*a^*b^*$ coordinates of the eight virgin olive oils studied: unfiltered samples (control sample) and filtered samples (cotton and paper plus anhydrous sodium sulphate) (\pm SD)

		L^*	a^*	b^*	Sample description
Sample 1	Control sample	63.2 \pm 0.0	6.7 \pm 0.0	99.0 \pm 0.2	Clean and deep green
	Cotton	65.6 \pm 0.0	6.5 \pm 0.0	97.8 \pm 0.1	
	Paper+SSA	65.5 \pm 0.1	6.8 \pm 0.0	101.8 \pm 0.2	
Sample 2	Control sample	63.2 \pm 0.2	3.8 \pm 0.0	88.1 \pm 0.6	Veiled and light green
	Cotton	70.7 \pm 0.1	1.9 \pm 0.1	93.4 \pm 0.2	
	Paper+SSA	70.1 \pm 0.3	1.8 \pm 0.0	92.7 \pm 0.4	
Sample 3	Control sample	61.1 \pm 0.1	1.1 \pm 0.0	59.7 \pm 0.0	Clean and light green
	Cotton	71.3 \pm 0.3	-1.5 \pm 0.1	62.9 \pm 0.1	
	Paper+SSA	74.0 \pm 0.1	-0.9 \pm 0.0	69.4 \pm 0.1	
Sample 4	Control sample	61.6 \pm 0.1	3.5 \pm 0.0	79.9 \pm 0.3	Veiled and light green
	Cotton	71.6 \pm 0.1	0.9 \pm 0.0	89.3 \pm 0.7	
	Paper+SSA	69.8 \pm 0.0	3.1 \pm 0.0	93.4 \pm 0.4	
Sample 5	Control sample	62.3 \pm 0.2	6.9 \pm 0.0	96.2 \pm 0.2	Very veiled and deep green
	Cotton	63.6 \pm 0.1	7.4 \pm 0.0	98.9 \pm 0.4	
	Paper+SSA	65.3 \pm 0.1	7.0 \pm 0.0	103.8 \pm 0.4	
Sample 6	Control sample	52.2 \pm 0.0	9.1 \pm 0.0	85.1 \pm 0.2	Very veiled and light green
	Cotton	54.3 \pm 0.0	8.4 \pm 0.0	86.1 \pm 0.1	
	Paper+SSA	54.2 \pm 0.1	8.5 \pm 0.0	87.8 \pm 0.2	
Sample 7	Control sample	67.2 \pm 0.4	6.2 \pm 0.1	101.5 \pm 0.4	Very veiled and deep green
	Cotton	67.8 \pm 0.1	6.9 \pm 0.0	103.8 \pm 0.5	
	Paper+SSA	68.9 \pm 0.0	7.0 \pm 0.0	104.8 \pm 0.5	
Sample 8	Control sample	60.3 \pm 0.2	4.1 \pm 0.1	85.5 \pm 0.7	Very veiled and light green
	Cotton	70.6 \pm 0.1	2.3 \pm 0.0	99.0 \pm 0.3	
	Paper+SSA	71.1 \pm 0.0	2.0 \pm 0.0	99.3 \pm 0.2	

Paper+SAA, filtration by paper and sodium sulphate anhydrous

sample had a deep green color, the a^* value increased after filtration and the intensity of green color was minimized; whereas if the control sample was light green, the a^* value decreased and the contribution of green color was more apparent. The b^* value had a tendency to increase because the yellow color was more evident when the olive oil had been filtered.

CONCLUSIONS

By study of two filtration systems it can be concluded that the oxidation stability decreases after filtration due to elimination of water. This could be due both to the decrease of the concentration of phenols with a higher antioxidant activity, particularly hydroxytyrosol and to the decrease of antioxidant activity of phenolic compounds when the water content is lowered. Furthermore, filtrated olive oils had a higher component of yellow color, luminosity and in some cases, the intensity of green color diminished. Presently, consumers have more knowledge about olive oil and would choose a veiled and deep green oil over one that is transparent and light green oil (filtered). However there are also consumers that prefer transparent oils. Thus filtration may reduce the quality of virgin olive oils (oxidative stability decrease) and many consumers may not prefer these products due to their unfavorable visual characteristics.

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