J Am Oil Chem Soc (2008) 85:253–262
DOI 10.1007/s11746-007-1186-6

ORIGINAL PAPER

Sterols, Fatty Alcohols, and Triterpenic Alcohols in Commercial Table Olives

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Received: 6 July 2007 / Revised: 5 November 2007 / Accepted: 30 November 2007 / Published online: 3 January 2008

Abstract This work supplies information on the lipids, unsaponifiable matter, sterols, and fatty and triterpenic alcohols in table olives. The mean lipid contents, unsaponifiable values, concentration of sterols and total alcohols (aliphatic and triterpenic alcohols) were 16.15 g/100 g edible portion (e.p.), 4.53 g/100 g lipid, 28.68 mg/100 g e.p. and 13.28 mg/100 g e.p., respectively. The overall mean content of cholesterol was 0.5 mg/100 g e.p., with a minimum of 0.08 mg/100 g e.p. in Manzanilla olives stuffed with "piquillo" pepper, and a maximum of 4.9 mg/100 g e.p. in Manzanilla olives stuffed with marinated anchovy strips. Table olives contain higher concentrations of phytosterols than olive oil. The chemometric analysis showed that lipids, unsaponifiable matter, sterols, and fatty and triterpenic alcohol contents in table olives were slightly affected by processing and that some misclassification was possibly related to maturation. There were also noticeable differences between cultivars.

Keywords Discriminant analysis · Fatty alcohols · Principal component analysis · Sterols · Table olives · Triterpenic alcohols

Introduction

Table olives constitute an important part of the Mediterranean diet and the world production reached a total of 1,700,000 tons in the 2003–2004 season. Spain produced and exported about 500,000 and 250,000 tons, respectively, during this period [1]. Table olives are restricted to only a few types or styles.

The most common are green olives (Spanish or Sevillian style), directly brined olives (turning color or naturally black olives), and ripe olives (California style). In brief, the procedure for preparing green Spanish-style olives consists of treating the fruits with a dilute NaOH solution, followed by water washes, and brining. In brine, olives undergo lactic acid fermentation. The commercial presentations of green olives are numerous and include the use of many stuffing materials [2]. Untreated olives (green, turning color or naturally black) are directly brined after picking, where they undergo a limited fermentation and lose some of their natural bitterness. According to market demand, olives are sorted, graded and packed. In some commercial presentations, they can be broken or cut along their higher longitudinal diameter and/or seasoned with natural products or their flavors. Olives for producing ripe olives (by alkaline oxidation) are previously preserved in an aqueous solution (brine, acidic water, etc.) and darkened throughout the year. Darkening consists of several treatments of dilute NaOH solutions and water washes, with aeration, between them. Darkened olives are immersed in a lactate or gluconate iron solution and packed in light brine. Their commercial presentations are limited to plain (whole), pitted, sliced, and, sometimes, olive paste [2].

Although investigations about the composition of olive oil are numerous, this is not the case in relation to composition of lipids from table olives. Their physiological benefits are probably determined by the large amount of minor components in the unsaponifiable fraction of olive oil. The amount of unsaponifiable matter is about 1% of the total lipid composition in olive oil [3]. The analysis of

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unsaponifiable components requires a collection via saponification, followed by thin layer chromatography (TLC) fractionation of the different classes present in the extract (hydrocarbons, carotenes, tocopherols, linear alcohols, triterpenic alcohols, methyl sterols, sterols and triterpenic dialcohols) [4].

Clinical studies have demonstrated that the dietary intake of plant sterols, also called phytosterols, as part of a normal diet, or as a supplement, may decrease blood cholesterol levels inhibiting its absorption from the small intestine [5]. Also, phytosterols have been recognized as cancer-preventive biologically active substances, although not yet confirmed in epidemiological studies [6]. Additionally, sterol and alcohol profiles are used to characterize virgin olive oils and especially to detect the adulteration of olive oil with hazelnut oil [7]. The presence of olive-pomace oil in virgin olive oil can be detected from the levels of triterpenic dialcohols (erythrodiol + uvaol), whose concentrations are considerably higher in pomace oils than in virgin and refined olive oils [8]. The sterol composition has been used to classify Portuguese olive oils by multivariate analysis according to the Protected Denomination of Origin [9]. Sterol and alcohol profiles have been proposed to classify virgin olive oils according to their fruit variety [10]. Chemometric studies have been used for the characterization of varietal olive oils based on their sterols and other fatty components (fatty acids, tocopherols, diacylglycerols, or triacylglycerols) [11].

The aim of this work was to determine the content of sterols, triterpenic dialcohols and fatty alcohols in table olives, taking into account cultivars, processing methods and commercial forms.

Experimental Procedures

Samples

Analyses were carried out in duplicate on composite samples from each commercial presentation, which were made up of 3–8 units (cans, jars or plastic pouches), depending on their sizes, and different packing dates, from 1 to 5 companies, according to their availability on market shelves. Producers [(Angel Camacho S.A. Morón de la Frontera (Sevilla), Jolca S.A. Tomares (Sevilla), and SCL del Campo San Marcos Almendralejo (Badajoz)] kindly supplied those commercial presentations not available in the local markets. Average time from packing was about 3 months. Samples belonged to the following styles, cultivars and commercial presentations.

Green Spanish-style. Gordal: plain, pitted, and seasoned. Gordal stuffed with: red pepper strips, natural red pepper, almond, cucumber, onions, garlic, and jalapeño. A blend of Gordal olives and red pepper strips called "salads". Manzanilla: plain, pitted, sliced, anchovy flavored, and plain seasoned. Manzanilla stuffed with: red pepper strips, anchovy strips, marinated anchovy strips, natural red pepper, almond, almond and red pepper, salmon strips, tuna strips, onions, capers, garlic, hazelnut, hot pepper, hot pepper strips, "piquillo" pepper, lemon paste, ham paste, orange strips, cheese, "jalapeño" strips, and garlic strips. A blend of pitted or slices of Manzanilla olives with red pepper strips called "pitted salads" and sliced "salads", respectively; a blend of Manzanilla olives with slices of carrot added called "gazpachas"; and a blend of Manzanilla olives and capers called "alcaparrado". Carrasqueña: pitted. A blend of pitted Carrasqueña olives and red pepper strips, called "salads"; and a blend of Carrasqueña olives and capers called "alcaparrado". Hojiblanca: plain, pitted, sliced. Hojiblanca olives stuffed with red pepper strips.

Directly brined olives. Gordal: broken "seasoned" turning color. Manzanilla: turning color in brine alone, "seasoned" turning color, and olives from biological (or ecological) production. Hojiblanca: "seasoned" turning color. Arbequina: "seasoned" turning color. Aloreña: green "seasoned" broken, prepared from fresh fruits and from stored olives. Verdial: green "seasoned" broken.

Ripe olives (by alkaline oxidation). Gordal: plain. Manzanilla: pitted. Carrasqueña: plain and pitted. Hojiblanca: plain, pitted, and sliced. Cacereña: plain, pitted, and sliced.

Determination of Lipids and Their Unsaponifiable Fraction

Lipids from the olives were obtained by extracting 12 g of lyophilized samples with hexane, in duplicate, for 6 h, using a Soxhlet apparatus. The solvent was removed in a rotary evaporator at 40 °C, and the residual oil was dried in an oven at 105 °C until a constant weight was reached [12].

The unsaponifiable matter was determined by saponification of the lipid with potassium hydroxide in ethanolic solution and extracted with diethyl ether [13].

Determination of Sterols and Triterpenic Dialcohols

This analysis was performed according to the method described by the Official Journal of the European Communities [14]. The lipids, with added α-cholestanol and betulin as internal standards, were saponified and the unsaponifiable matter extracted as mentioned above. The bands corresponding to the sterols and triterpene alcohols fractions were separated from the extract by TLC on a basic silica gel plate. The sterols and erythrodiol and uvaol

recovered from the plate were transformed into trimethylsilyl ethers and the mixture was analyzed by GC using an HP 5890 Series II gas chromatograph equipped with a flame ionization detector and a 30 m × 0.32 mm i.d. Tracsil TRB-5 (95% dimethylpolysiloxane-5% diphenyl, film thickness 0.25 μm) capillary column (Teknokroma, Barcelona, Spain). The chromatographic conditions were: injector 300 °C, isothermal column 275 °C, and detector 300 °C. Split ratio was 1:50. Hydrogen carrier gas was used at 1.0 mL/min.

Determination of Fatty Alcohols

This analysis was performed according to the method described by the Official Journal of the European Communities [15]. The fatty substance, with 1-eicosanol added as internal standard, was treated as mentioned in the determination of the lipid fraction and unsaponifiable fraction section. The alcohol fraction was separated from the unsaponifiable matter by chromatography on a basic silica gel plate. The alcohols recovered from the silica gel were transformed into trimethylsilyl ethers and analysed using capillary gas chromatography. The chromatographic conditions were the same as those mentioned above for sterols and triterpenic dialcohols, except that oven temperature was as follows: 215 °C (5 min); 3 °C/min ramp to 290 °C; and 2 min hold. All analyses were performed in duplicate.

Statistical Analyses

Data from the analyses were arranged in a 134 × 17 matrix array, where rows were cases and columns were variables (total lipids, unsaponifiable fraction, sterols, and fatty and triterpenic alcohols). Data from brassicasterol, Δ^7-stigmastenol, and Δ^7-avenasterol were discarded because they were not detected in most of the samples or were found in low concentrations close to the quantification limits.

Data were standardized before being subjected to the chemometric analysis and were successively studied by multiple analysis of variance (MANOVA) to test overall differences between groups across the different variables, principal component analysis (PCA), and discriminant analysis (DA). PCA was carried out, using a varimax rotation, to detect the data structure and to determine the relationship between samples and original variables. For the selection of the number of principal components (PCs), the Kaiser criterion [16] was followed and only factors with eigenvalues (e.v.) higher than 1.00 were retained. Then, the loadings of the original variables were projected onto the factorial plane formed by the first and second component.

The selection of variables containing the most powerful information for correct classification of olive samples of the three (types) or eight (cultivars) categories, was carried out on the basis of the canonical analysis of data, using backward stepwise analysis option, which first includes all the variables in the model and then, at each step, eliminates the variable that least contributes to membership prediction. The process continues until only the important variables that contribute most to discrimination between groups are in the model. The values of probability to enter or to remove were fixed at 0.05 and 0.1, respectively. The number of steps was fixed at 100, the minimum tolerance at 0.001 and no variable was forced to enter in any model. The scores of table olive samples were plotted on the canonical axes (discriminant coordinates, called Factors). These axes were determined in such a way that the rate of the variance between groups to the variance intra-groups are maximized [17].

Classification was achieved by means of the corresponding classification functions. For k groups, k linear combinations of variables are constructed, called classification functions. The calculation of the values of these functions for each sample makes it possible to allocate this sample to the group for which the probability of belonging is the highest. The prior probabilities were established in proportion to the number of samples in each group.

A leaving-one-out cross validation procedure was performed for assessing the performance of the classification rule. In this last step, the sample data minus one observation was used for the estimation of the classification functions, and then the omitted variable was classified from them. The procedure was repeated for all samples. Consequently, each sample was classified by classification functions which were estimated without its contribution [17].

The different statistical techniques used in this work were implemented using STATISTICA, release 6.0 and SYSTAT, release 10.2.

Results and Discussion

The frequency distributions (histograms) of the contents of lipids, unsaponifiable fraction, the total sterol, and total alcohols in all samples of table olives are shown in Fig. 1. The highest proportion of the table olive samples (about 60%) had between 14 and 20 g total lipids/100 g edible portion (e.p.). The mean content was 16.15 g/100 g e.p, and varied between 8.73 (Gordal olives stuffed with cucumber) and 31.07 g/100 g e.p. (Arbequina olives seasoned turning color). In the case of unsaponifiable fraction, the highest percentage of samples (about 55%) was in the range 2–5 g/100 g lipid. The mean value was 4.53 g/100 g

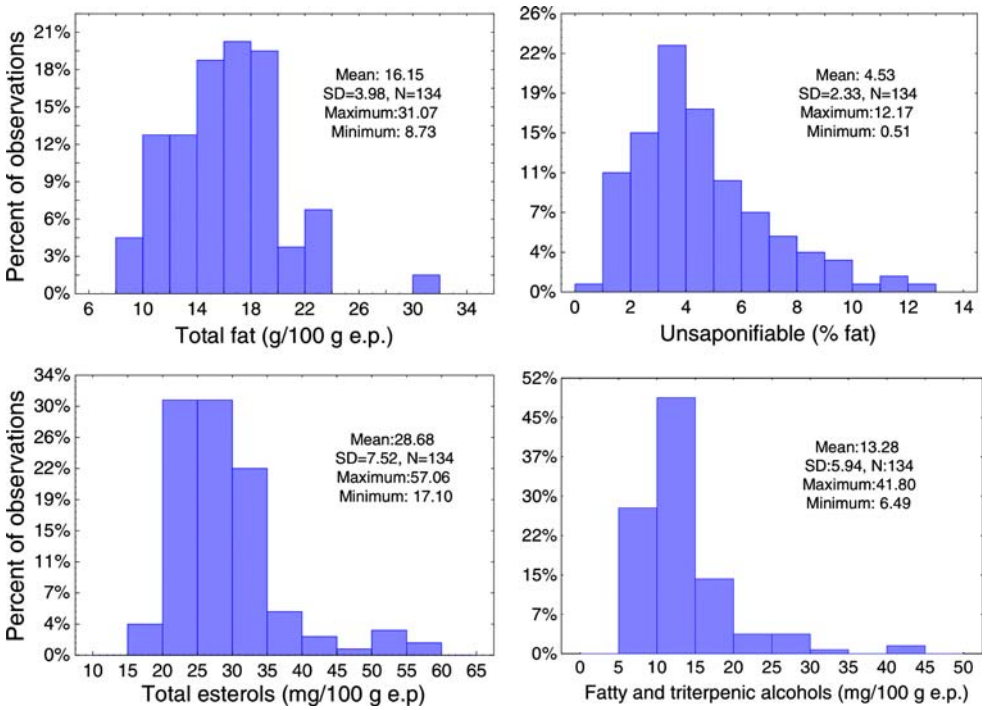

Fig. 1 Distribution of total lipids (g/100 g e.p), total unsaponifiable fraction (% lipids), total sterols (mg/100 g e.p), and fatty and triterpenic alcohols (mg/100 g e.p.) in the main commercial presentations of table olives

lipids, with a minimum of 0.51 g/100 g lipids in seasoned Arbequina turning color olives, and a maximum of 12.17 g/100 g lipids in pitted Carrasqueña olives. This mean value is higher than that usually found in olive oil ($\approx$1%) [3]. Most commercial samples (around 80%) had a total sterol content of 20–35 mg/100 g e.p. The mean concentration was 28.68 mg/100 g e.p. with a minimum of 17.10 mg/100 g e.p. in sliced Cacereña olives, and a maximum of 57.06 mg/100 g e.p. in seasoned Arbequina turning color olives. With respect to total alcohols, including both aliphatic and triterpenic alcohols, the mean concentration was 13.28 mg/100 g e.p. with a minimum of 6.49 mg/100 g e.p. in Gordal olives "salads", and a maximum of 41.80 mg/100 g e.p. in seasoned Arbequina turning color olives.

A comparison between processing types demonstrated that there was a statistical difference ($p < 0.05$) in their contents of lipids, the unsaponifiable fraction, total sterols, and total alcohols (Fig. 2). The highest lipid content as well as the highest content of total sterols and total alcohols was found in directly brined olives (mean values of 21.8, 39.5, and 24.8 mg/100 g e.p, respectively). In contrast, samples in this processing type had the lowest content of unsaponifiable fraction (mean 1.9 g/100 g lipids). Unsaponifiable content could be related to the severity of changes suffered by olives depending on processing steps (e.g. alkaline treatment, oxidizing step, heat treatment). Thus, samples of ripe olives had the highest content of unsaponifiable fraction (mean 7.6 g/100 g lipids).

The mean concentration of sterols was 28.68 mg/100 g e.p. with a minimum of 17.10 mg/100 g e.p. in Cacereña sliced olives, and a maximum of 57.06 mg/100 g e.p. in seasoned Arbequina turning color olives. Taking into account the sterol composition in virgin olive oil [9], the major sterols were β-sitosterol, Δ^5-avenasterol and campesterol, with overall mean contents of 23.5, 1.5, and 0.9 mg/100 g e.p., respectively (Table 1). Brassicasterol, Δ^7-stigmastenol, and Δ^7-avenasterol were not detected. Mean values of each sterol were significantly ($p < 0.05$) higher in samples of directly brined olives compared to the other processing types, with the exception of cholesterol and stigmasterol. In green olives, the mean concentrations of β-sitosterol, campesterol, and campestanol found in the present study were slightly lower than those reported in the literature (34, 1.1, and 0.09 mg/100 g e.p., respectively; [18]) but the concentration of stigmasterol was higher (0.51 vs. 0.29 mg/100 g e.p.).

Most lipids of table olives (90%) had cholesterol contents of 0–0.5 mg/100 g e.p. The overall mean content of cholesterol was 0.5 mg/100 g e.p., with a minimum of 0.08 mg/100 g e.p. in Manzanilla olives stuffed with "piquillo" pepper, and a maximum of 4.9 mg/100 g e.p. in Manzanilla olives stuffed with marinated anchovy strips. Other commercial samples with relatively high cholesterol contents were 3.4 mg/100 g e.p. in Manzanilla olives stuffed with anchovy strips, 2.6 mg/100 g e.p. in Manzanilla olives stuffed with salmon strips, 1.7 mg/100 g e.p. in Manzanilla stuffed with ham paste, and 1.1 mg/100 g e.p.

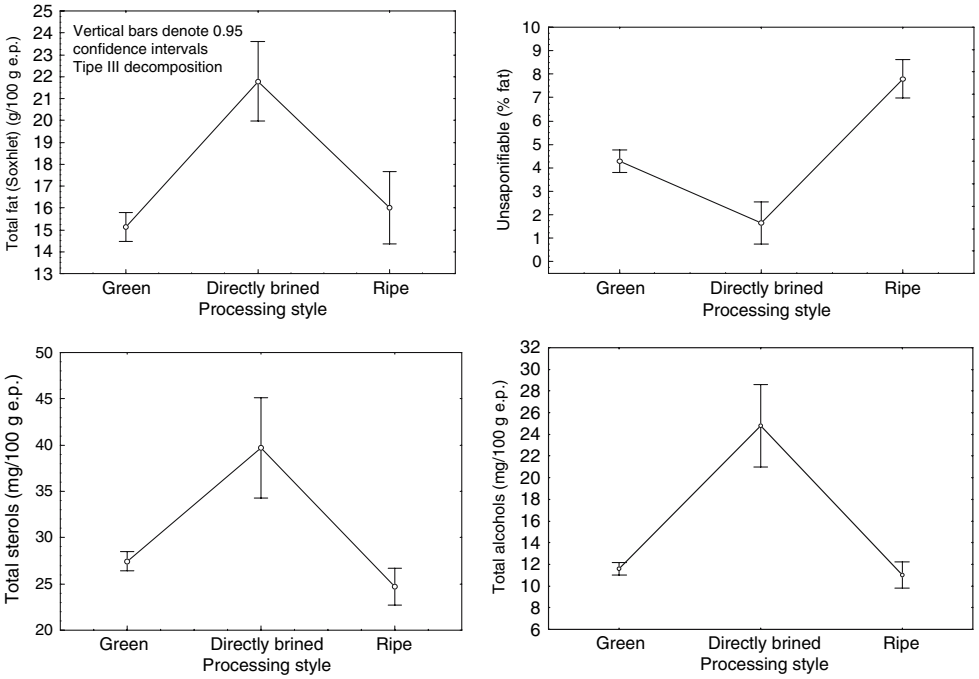

Fig. 2 Total lipids (g/100 g e.p), unsaponifiable matter (% lipid), total sterols (mg/100 g e.p.) and total (fatty and triterpenic) alcohols (mg/100 g e.p.) contents in commercial presentations of table olives, according to processing styles

Table 1 Mean values (mg/100 g e.p., standard error in parentheses) of the contents of sterols and alcohols in the different processing types of table olives studied

Composition	Processing style			
	Green olives (n = 48)	Directly brined olives (n = 9)	Ripe olives (n = 10)	Mean (n = 67)
Sterols				
Cholesterol	0.52 (0.09) a	0.31 (0.07) a	0.26 (0.02) a	0.45 (0.75)
Campesterol	0.86 (0.02) a	1.30 (0.10) b	0.70 (0.03) a	0.89 (0.29)
Campestanol	0.08 (0.01) a	0.17 (0.03) b	0.09 (0.02) a	0.10 (0.10)
Stigmasterol	0.51 (0.05) a	0.61 (0.04) a	0.33 (0.02) a	0.49 (0.43)
Δ^7-Campesterol	0.55 (0.04) ab	0.91 (0.15) b	0.32 (0.08) a	0.56 (0.51)
Clerosterol	0.79 (0.04) a	1.28 (0.19) b	0.61 (0.05) a	0.83 (0.51)
β-Sitosterol	22.51 (0.41) a	31.83 (2.16) b	20.85 (0.87) a	23.51 (5.98)
Δ^5-Avenasterol	1.28 (0.05) a	2.78 (0.48) b	1.33 (0.10) a	1.49 (1.00)
$\Delta^{5,24}$-Stigmastadienol	0.19 (0.02) a	0.45 (0.15) b	0.12 (0.03) a	0.21 (0.31)
Alcohols				
Dicosanol	0.45 (0.01) a	1.25 (0.17) b	0.35 (0.03) a	0.54 (0.41)
Tetracosanol	0.95 (0.05) a	2.92 (0.40) b	0.64 (0.08) a	1.17 (1.04)
Hexacosanol	2.96 (0.10) a	7.73 (0.85) b	2.39 (0.22) a	3.52 (2.31)
Octacosanol	4.17 (0.11) a	7.45 (0.56) b	3.63 (0.10) a	4.73 (1.54)
Erythrodiol	2.25 (0.06) a	4.61 (0.57) b	2.64 (0.40) a	2.63 (1.44)
Uvaol	0.82 (0.06) a	0.85 (0.18) a	1.36 (0.30) a	0.91 (0.76)

Means in a row with different letters are significantly different ($p < 0.05$)

n Number of commercial presentations

in Manzanilla olives stuffed with tuna strips. Bearing in mind that the serving size of olives for USA and Canada is around 15 g and that this nutrient must be declared only if its concentration is above 2 mg in this portion, cholesterol can be always declared as 0, even in those commercial products with the highest contents.

With respect to total alcohols, including both aliphatic and triterpenic alcohols, the mean concentration was 13.28 mg/100 g e.p. with a minimum of 6.49 mg/100 g e.p. in Gordal olives "salads", and maximum 41.80 mg/100 g e.p. in seasoned Arbequina turning color olives. Octacosanol, hexacosanol, and erythrodiol were the most abundant, with overall mean values of 4.7, 3.5, and 2.6 mg/100 g e.p., respectively. Again, the highest contents of individual alcohols were found in directly brined olives, with the exception of uvaol (Table 1). Concentrations of sterols and triterpenic dialcohols are higher in olive-pomace oils (which are extracted from olive pomace with solvents) than in virgin and refined olive oils [19]. When the mean contents of sterols and alcohols in table olives were expressed as mg/100 g total lipid (using the mean contents of total lipids for each processing type) and compared with the reported contents in olive oils, it was observed that in general values of individual sterols and alcohols were higher than those of virgin olive oil but lower than those of pomace olive oil (data not shown).

Comparing sterol contents between cultivars for each processing type (Table 2), it was found that significant ($p < 0.05$) differences between cultivars occurred for several sterols in samples of directly brined olives, with Arbequina and Aloreña cultivars showing the highest contents of total sterols and β-sitosterol. These cultivars along with Verdial cultivar had also the highest values of campesterol and Δ^5-avenasterol.

In green or ripe olives no significant differences between cultivars were found in any case. It must be noted that the cholesterol level for the Manzanilla cultivar in samples of green olives was relatively high in comparison with the other cultivars, but differences were not statistically different. It is explained by the great variability in cholesterol content as result of the high number of different lipids within the Manzanilla cultivar.

Significant differences among cultivars with regards to contents of triterpenic and fatty alcohols were found in both green and directly brined olives (Table 3). Such differences were small in green olives. In directly brined olives, Arbequina cv. had the highest content of total fatty alcohols as well as individual fatty alcohols; Verdial cv. showed the highest level of erythrodiol as well as total triterpenic alcohols. In ripe olives only erythrodiol content showed significant differences among cultivars (Manzanilla and Carrasqueña cultivars had the highest values).

The multivariate analyses of the standardized data showed that cholesterol and Δ^7-campesterol were the only compounds which showed no significant differences among elaboration types ($p = 0.429$ and 0.108, respectively) and cultivars ($p = 0.453$ and 0.099, respectively). The data were then subjected to the PCA. Only 5 e.v. higher than 1.00 were obtained. Their values (and the percentage of total variance explained) were; 6.48 (38.09%), 1.91 (11.23%), 1.32 (7.77%), 1.15 (6.76%), 1.07 (6.27%). The cumulative explained variance by the 5 e.v. was 70.11%. The following PCs progressively explained less and less variance. The loadings associated to each variable on these five principal components (PCs) identified the variables that most contribute to them. PC1 was associated with all variables except cholesterol and uvaol. PC2 was related to $\Delta^{5,24}$-stigmastadienol, Δ^5-avenasterol, dicosanol (C24), hexacosanol (C26), octacosanol (C28), tetracosanol (C24), Δ^7-campesterol, campestanol, stigmasterol, and erythrodiol. The projections of the loadings on the plane of the two first PCs are illustrated in Fig. 3. These projections allow visualizing the position of the variables in the plane and the corresponding correlations. Two distant variables (the angle between the corresponding variables is 90°) are not correlated because the correlation coefficient is the cosine of this angle (0, in the example) [20]. Figure 3 shows that the fatty alcohols are highly correlated but the correlation between uvaol and erythrodiol (triterpenic alcohols) was lower. However, correlation between sterols was diverse and especially cholesterol had a low correlation with any of them. Apparently, there was good agreement among total lipid and most of the sterols and alcohols, and a negative relationship among all of them and the percentage of unsaponifiable. When the scores for the cases were plotted as a function of PC1 versus PC2 no evident segregation among cases according to treatments and cultivars was observed (data not shown). Then predictive DA was applied to the standardized data matrix because this analysis is a more powerful tool for classification. The selection of variables containing the most powerful information for correct classification of olives was achieved by estimating the canonical discriminant functions. The most discriminant variables among elaboration styles were: total lipid (F to remove, 4.02), campesterol (6.38), dicosanol (C22) (10.65), hexacosanol (C26) (3.32), erythrodiol (14.71), and uvaol (5.43). Therefore, fatty and triterpenic alcohols play an important role in discriminating among styles. The procedure also determines the corresponding canonical variables. Plotting the loads of the samples on the plane defined by the two canonical variables makes it possible to visualize the differences among the groups (Fig. 4). Application of the classification functions led to the confusion matrix shown in Table 4. Overall correct answers were 83%; however, the response depended on the elaboration type. The classification efficiency was fairly good in green olives but was low (20%) in ripe olives because most of the samples were classified as green. Directly brined olives had 72%

Table 2 Mean values (mg/100 g e.p.) of the contents of sterols in commercial table olives according to olive cultivars

Cultivar	Cholesterol	Campesterol	Campestanol	Stigmasterol	Δ^7-Campesterol	Clerosterol	β-Sitosterol	Δ^5-Avenasterol	$\Delta^{5,24}$-Stigmastadienol	Total sterols
Green olives										
Gordal	0.32 a	0.87a	0.06 a	0.38 a	0.74 a	0.95 a	21.70 a	1.23 a	0.24 a	25.94 a
Manzanilla	0.66 a	0.87 a	0.10 a	0.58 a	0.48 a	0.72 a	22.55 a	1.31 a	0.18 a	27.53 a
Carrasqueña	0.25 a	0.77 a	0.08 a	0.40 a	0.61 a	0.78 a	21.28 a	1.47 a	0.19 a	25.85 a
Hojiblanca	0.23 a	0.82 a	0.06 a	0.35 a	0.44 a	0.84 a	25.30 a	1.04 a	0.10 a	29.21 a
Directly brined olives										
Gordal	0.18 a	0.94 a	0.20 a	0.55 a	0.92 ab	0.81 ab	28.29 a	0.89 a	ND	32.78 a
Manzanilla	0.29 a	1.04 a	0.13 a	0.65 a	0.96 ab	1.48 ab	28.07 a	1.45 a	0.34 a	34.60 a
Hojiblanca	0.14 a	0.93 a	0.24 a	0.35 a	0.24 a	0.42 a	23.78 a	1.69 a	ND	27.79 a
Arbequina	0.19 a	1.97 b	0.17 a	0.64 a	1.12 ab	1.03 ab	43.53 b	4.87 b	0.64 ab	54.78 b
Aloreña	0.27 a	1.62 b	0.11 a	0.50 a	0.44 a	2.15 b	43.01 b	4.00 b	0.50 ab	52.63 b
Verdial	0.24 a	1.52 ab	0.33 a	1.01 a	2.17 b	0.51 a	20.66 a	5.24 b	1.38 b	33.05 a
Ripe olives										
Gordal	0.24 a	0.77 a	0.15 a	0.38 a	0.17 a	0.44 a	19.69 a	0.66 a	0.07a	22.57 a
Manzanilla	0.19 a	0.77a	ND	0.46 a	0.55 a	0.74 a	26.13 a	1.26 a	ND	30.10 a
Carrasqueña	0.36 a	0.84 a	0.20 a	0.45 a	0.54 a	0.80 a	22.45 a	1.85 a	0.24 a	28.01 a
Hojiblanca	0.25 a	0.70 a	0.05 a	0.28 a	0.30 a	0.60 a	22.63 a	1.04 a	0.05 a	25.95 a
Cacereña	0.22 a	0.57 a	0.05 a	0.23 a	0.16 a	0.52 a	16.62 a	1.53 a	0.16 a	20.22 a

Mean values with different letters within a column for each processing type are significantly different ($p < 0.05$)

ND Not detected

Table 3 Mean values (mg/100 g e.p.) of the contents of triterpenic and fatty alcohols in commercial table olives according to olive cultivars

Cultivar	Erythrodiol	Uvaol	Dicosanol	Tetracosanol	Hexacosanol	Octacosanol	Erythrodiol + Uvaol	Total fatty alcohols
Green olives								
Gordal	1.61a	0.59 a	0.37 a	0.35 a	1.98 a	4.02 a	2.19 a	6.70 a
Manzanilla	2.33 b	0.76 a	0.49 a	1.21 b	3.20 b	4.19 a	3.09 ab	9.09 b
Carrasqueña	3.02 b	0.87 ab	0.42 a	0.94 ab	3.26 ab	3.92 a	3.89 ab	8.53 ab
Hojiblanca	2.83 b	1.92 b	0.39 a	0.66 ab	3.65 b	4.60 a	4.75 b	9.29 ab
Directly brined olives								
Gordal	2.37 ab	0.27 a	0.82 ab	1.71 a	5.24 ab	6.52 ab	2.63 ab	14.28 ab
Manzanilla	3.35 ab	0.95 a	1.63 c	3.74 b	9.05 c	7.59 ab	4.30 ab	22.01 c
Hojiblanca	1.89 a	0.40 a	1.26 bc	2.73 ab	3.84 a	5.10 ab	2.29 a	12.93 ab
Arbequina	4.53 bc	1.28 a	2.55 d	6.28 c	15.01 d	12.15 c	5.81 bc	35.99 d
Aloreña	6.54 c	0.39 a	0.55 a	1.31 a	7.83 bc	8.21 b	6.93 c	17.89 bc
Verdial	9.52 d	2.08 a	0.63 a	1.76 a	2.64 a	4.13 a	11.59 d	9.16 a
Ripe olives								
Gordal	1.46 a	1.15 a	0.26 a	0.15 a	0.75 a	3.45 a	2.61 a	4.61 a
Manzanilla	3.05 ab	ND	0.42 a	1.29 a	3.23 a	3.94 a	3.05 a	8.87 a
Carrasqueña	4.72 b	0.82 a	0.35 a	0.87 a	2.72 a	3.67 a	5.54 a	7.62 a
Hojiblanca	2.08 a	2.11 a	0.36 a	0.52 a	2.01 a	3.29 a	4.19 a	6.16 a
Cacereña	2.07 a	1.51 a	0.35 a	0.55 a	2.83 a	3.89 a	3.58 a	7.61 a

ND Not detected

Mean values with different letters within a column for each processing type are significantly different ($p < 0.05$)

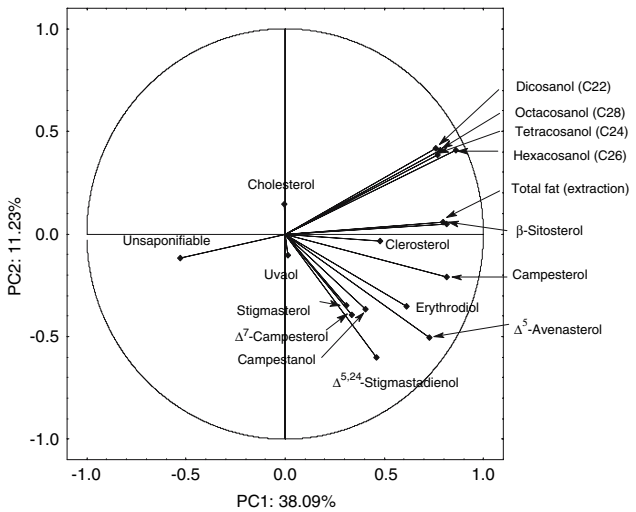

Fig. 3 Projection (loadings) of the variables in the first two principal components

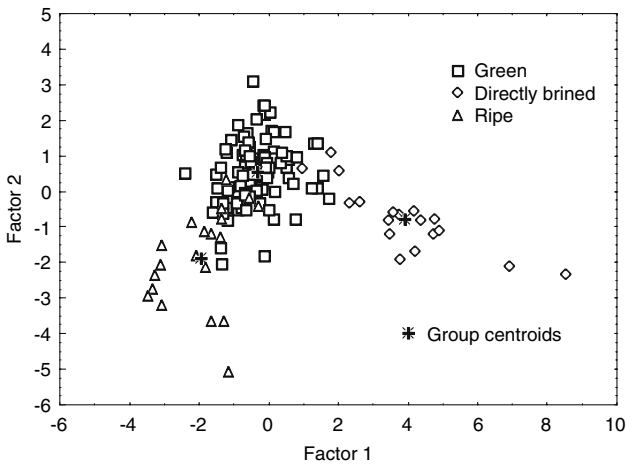

Fig. 4 Plot of table olive commercial presentation sample scores as a function of the two canonical discriminant functions, according to processing styles

correct assignations. Cross-validation yielded similar results. The failure to correctly assign ripe olives to its group may be due to the observations that the content in total lipids, unsaponifiable fraction, sterols, and fatty and triterpenic alcohols are strongly related to the maturation degree and assign ripe olives to green olives because they are picked at similar maturation stages. The partial failure in directly brined olives may be related to the fact that these are a mixture of samples with different maturation stages (from green to naturally black).

Table 4 Classification matrix according to processing styles (Jackknifed classification in parenthesis)

Actual group	Predicted group membership			
	Green	Directly brined	Ripe	% Correct
Green	94 (94)	0 (0)	2 (2)	98 (98)
Directly brined olives	5 (5)	13 (13)	0 (0)	72 (72)
Ripe	16 (16)	0 (1)	4 (3)	20 (20)
Total	115	13	6	83 (82)

The most contributing variables for discrimination among cultivars were: total lipids (F to remove, 12.44), campesterol (3.91), campestanol (3.53), Δ^7-campesterol (4.00), β-sitosterol (14.98), Δ^5-avenasterol (9.37), tetracosanol (6.84), hexacosanol (5.26), erythrodiol (32.11), and uvaol (10.71). The most outstanding contributions came from the triterpenic alcohols. Visualizing the discrimination among variables as a function of the loads of the samples in each canonical factor is difficult because of the high number of cultivars studied; however, the overall classification obtained was higher than among elaboration styles (89%) (Table 5). Values of cross validation were slightly lower, except in the case of Verdial. In any case, the classification of cultivars with a low number of cases must be taken with precaution due to the limitations derived from the application of the DA to a reduced number of samples. The best discriminated cultivars were Arbequina, Aloreña, Manzanilla, Gordal, and Cacereña. Hojiblanca and Carrasqueña were hardly distinguished because of their classification as Manzanilla. This is reasonable for Carrasqueña because it is very similar to Manzanilla and their differences are usually attributed to the diverse characteristics of the growing areas. However, the difficulties to distinguish Hojiblanca from Manzanilla may favor the use of one over the other and facilitate possible frauds. The higher capacity of the variables studied to distinguish among cultivars than among elaboration styles may indicate that these compounds are only slightly or not at all affected by processing conditions. The chemometric analysis has shown that lipid, unsaponifiable matter, sterols, and fatty and triterpenic alcohol contents in table olives was slightly affected by processing and that some misclassification were possibly related to maturation. There were also noticeable differences among cultivars.

Information on lipids, unsaponifiable matter, sterols, and fatty and triterpenic alcohols in table olives was scarce. This work has studied in detail their contents in the main commercial presentations of Spanish table olives. After water, the lipids are the main constituents of table olives which concentrations ranged from 8 to 24 g/100 g e.p. and the unsaponifiable proportion in them was from 0 to 13%. Overall, table olives contained higher concentrations of phytosterols than olive oil; sterols were the most abundant components of the unsaponifiable (15–60 mg/100 g e.p.) followed by fatty and triterpenic alcohols (5–35 mg/100 g e.p.). Cholesterol content in table olive was fairly low (average 0.5 mg/100 g e.p.). The minimum level (0.08 mg/100 g e.p.) was found in Manzanilla olives stuffed with "piquillo" pepper and the maximum (4.9 mg/100 g e.p.) in Manzanilla olives stuffed with marinated anchovy strips. Lipid, unsaponifiable matter, sterols, and fatty and triterpenic alcohol contents in table olives were slightly affected by processing as demonstrated by chemometric analysis, which showed noticeable differences between cultivars. However, the effect due to maturation was low and led to some misclassifications.

Table 5 Classification matrix according to cultivars (Jackknifed classification in parenthesis)

Actual group	Predicted group membership								
	Gordal	Manzanilla	Carrasqueña	Hojiblanca	Arbequina	Aloreña	Verdial	Cacereña	% Correct
Gordal	24 (24)	2 (2)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	92 (92)
Manzanilla	0 (0)	69 (69)	0 (0)	1 (1)	0 (0)	0 (0)	0 (0)	0 (0)	99 (99)
Carrasqueña	2 (2)	3 (3)	4 (3)	0 (1)	0 (0)	0 (0)	1 (1)	0 (0)	40 (30)
Hojiblanca	0 (1)	5 (5)	0 (0)	9 (8)	0 (0)	0 (0)	0 (0)	0 (0)	64 (57)
Arbequina	0 (0)	0 (0)	0 (0)	0 (0)	2 (2)	0 (0)	0 (0)	0 (0)	100 (100)
Aloreña	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	4 (4)	0 (0)	0 (0)	100 (100)
Verdial	0 (0)	0 (0)	0 (1)	0 (0)	0 (0)	0 (0)	2 (1)	0 (0)	100 (50)
Cacereña	0 (0)	1 (2)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	5 (4)	83 (67)
Total	26 (27)	80 (81)	4 (4)	10 (10)	2 (2)	4 (4)	3 (2)	5 (4)	89 (86)

Acknowledgments This work was supported by the Spanish Government (AGL-2003-00779 and AGL-2006-03540/ALI), European Union (TDC-Olive) and Junta de Andalucía (throughout financial support to group AGR-125). We also thank Amparo Cortés for technical assistance.

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