JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY

Multivariate Analysis for the Evaluation of Fiber, Sugars, and Organic Acids in Commercial Presentations of Table Olives

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Table olives constitute an important part of the Mediterranean diet and the diet of many non-olive-producing countries. The aim of this work was to determine the fiber, sugar, and organic acid contents in Spanish commercial presentations of table olives and characterize them by means of a multivariate analysis. The selection of variables was carried out on the basis of a canonical analysis and their classification, according to processing styles and cultivars, through a linear discriminant analysis. Values of dietary fiber in table olives ranged from 2 to 5 g/100 g edible portion (e.p.). Some stuffing materials (almond, hot red pepper, and hazelnut) or the addition of capers produced a significant increase in the total dietary fiber in green olives. Glucose, fructose, and mannitol were usually found in the ranges of 0–55, 0–70, and 0–107 mg/100 g e.p., respectively. Succinic acid was detected only in green and directly brined olives (0–40 mg/100 g e.p.), while lactic and acetic acids were used within the ranges of 0–681 and 5–492.8 mg/100 g e.p., respectively. A multivariate analysis showed that fiber, mannitol, and succinic, lactic, and acetic acids can be used to discriminate between processing styles (95.5% correct assignations) and cultivars (61.20%). Current data can also be used in the evaluation of the dietary value of table olives.

KEYWORDS: Table olives; dietary fiber; sugars; organic acids; glucose; fructose; succinic acid; lactic acid; acetic acid; multivariate analysis.

INTRODUCTION

Table olives are the main fermented product in western countries (1,700,000 tonnes in the 2003/2004 season) with a cumulative 2–5% increase every year (*1*). Table olives constitute an important part of the Mediterranean diet and the diet of many nonproducing countries around the world. Industrial elaborations are limited to only a few styles: green Spanish style, directly brined (turning color or naturally black), and ripe olives (darkened by oxidation). Green Spanish style olives are prepared by treating the fruits with a dilute NaOH solution (lye), followed by water washings, to remove the excess of alkali, and brining, where olives undergo lactic acid fermentation. Then, fruits are graded, sorted, conditioned, and packed according to diverse commercial presentations. Directly brined olives are immersed in brine just after picking, where they partially lose their natural bitterness. Then, olives are sorted, graded, and packed. Sometimes, they are cracked or incised and/or seasoned with natural products or their flavors. Ripe olives are previously stored in brine or in an acidic solution, darkened by oxidation, and, finally, packed in light brine. Their commercial presentations are limited to plain (whole), pitted, sliced, and, sometimes, olive paste. Cultivars and processing may affect the composition of table olive commercial presentations due to the different treatments applied and the diverse conditioning operations (pitting, slicing, and stuffing) (*2*).

Consumers are aware of the relationship between nutrition and health, and they demand detailed and updated information about food components. Among these are fiber, sugars, and organic acids. Trowell (*3*) defined dietary fiber as "the total polysaccharide and lignin of the diet that is not digested by endogenous secretions of the digestive tract", but this interpretation has changed according to the scientific field. The relevance of this food component to human health is free of controversy. Fiber intake can decrease the incidence of several diseases like cardiovascular complications, cancer (colon, breast, and prostate), and hypercholesterolemia (*4*). The cell walls of fruits, vegetables, pulses, and cereals make up the bulk of dietary fiber intake (*5*). Studies on the different fractions of dietary fiber and cell wall characterization in olives have been carried out for fresh (*6*) and processed (*7*) cultivars. The total crude fiber content was determined by De Castro Ramos et al. (*8*) in different olive laboratory scale samples. Food Composition and Nutrition Tables (*9*) contain limited data on table olive fiber. Sugars (mainly glucose, fructose, and mannitol) as well as organic acids are present in fresh olive fruits (*2*) and are

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10.1021/jf0720638 CCC: $37.00
Published on Web 12/01/2007

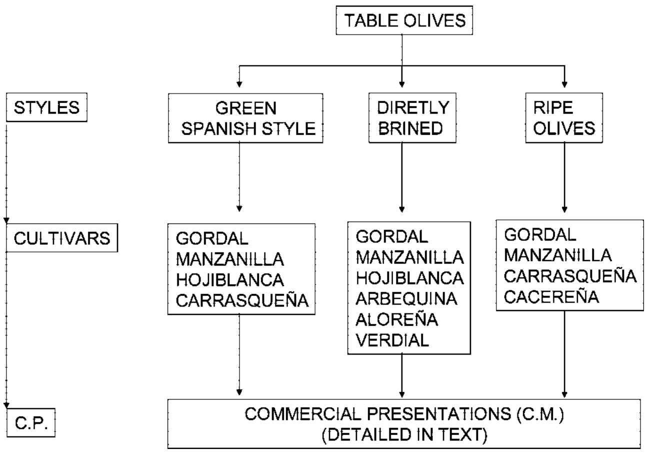

Figure 1. Scheme of the experimental design. A detailed description of the commercial presentations can be found in the text (Samples).

transformed by lactic acid bacteria and yeasts into lactic acid, acetic acid, and other volatile compounds during olive fermentation or storage; sugar consumption is not always complete and may pass partially into the packed products especially in seasoned or directly brined olives. Lactic and/or acetic acids, in combination or not with citric and/or ascorbic acids, are also used in the cover brines of olive packing. Ripe olives are stored in lactic or acetic acid solutions. These acids are used to control pH in the final product (*2*).

A multivariate analysis has been successfully used for evaluation of the lactic acid content in fermented cabbage juices (*10*) and vegetables juices (*11*) or the investigation of the origin of heptadecenoic and conjugated linolenic acids in milk (*12*). It has also been recently used to characterize table olives on the basis of their fatty acid composition (*13*).

The aim of this work was to determine the fiber, sugar, and organic acid contents in the Spanish commercial presentations of table olives and to use these compositions for the multivariate characterization according to processing styles and cultivars.

MATERIALS AND METHODS

Sample. Analyses were carried out on composite samples from the diverse commercial presentations. A scheme of the experimental design is shown in **Figure 1**. Each sample was made up of three to eight units (cans, jars, or plastic pouches), depending on their size, and different packing dates, from one to five elaboration companies, according to their availability on market shelves. Commercial presentations not available in the local markets were supplied by several producers and treated similarly. Average time from packing was approximately 3 months. All samples were of Spanish origin and 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 and sliced 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 biologic (or organic) 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. Gordal: plain. Manzanilla: pitted. Carrasqueña: plain and pitted. Hojiblanca: plain, pitted, and sliced. Cacereña: plain, pitted, and sliced.

Physicochemical Determinations. *Moisture Determination.* Olives were pitted when necessary, and their flesh was homogenized. Three aliquots of each sample (25 g) were weighed to within 0.1 mg and freeze-dried (Laboratory Freeze-Dryer, Telstar Cryodos) until a constant weight was reached.

Fat Extraction. Fat content was determined from dried samples in triplicate by Soxhlet extraction with hexane for 6 h. The solvent was removed in a rotary evaporator at 40 °C, and the residual oil was dried.

Protein Determination. The protein content was estimated by the micro Kjeldahl method (Büchi, Distillation Unit K-314) on the residue of one of the replicates after fiber determination. Kjeldahl nitrogen was converted into protein using a factor of 6.25.

Ash Determination. Another replicate of fiber determination was reduced to white ashes in an oven at 450 °C overnight.

Dietary Fiber Determination. Four replicates of dry defatted pulp were dispersed in a buffer solution of MES/TRIS as described by Lee et al. (*14*) and sequentially treated with the enzymes (a) α-amylase (100 °C, 15 min), (b) protease (60 °C, 30 min), and (c) amyloglucosidase (60 °C, 30 min). After digestion, hot (60 °C) ethanol was added to precipitate the soluble fiber; however, the precipitate was never obtained. Thus, total and insoluble fiber was the same. Ash and protein corrections were made at this step. Total dietary fiber content was then expressed as grams per 100 g of edible portion (e.p.), taking into account the moisture, fat, and ash contents.

Sugar and Organic Acid Determinations. Carbohydrates (sucrose, glucose, fructose, and mannitol) were assessed using a Hewlett-Packard series 1050 liquid chromatograph equipped with a Rheodyne 7125 injector and a column heater, a Perkin-Elmer model LC-25 refractive index detector, and a Hewlett-Packard model 3396 series II integrator. An Aminex HPX-87C column [300 mm × 7.8 mm (inside diameter), Bio-Rad Laboratories] held at 65 °C and deionized water as the eluent at a rate of 0.7 mL/min were used for the analysis of carbohydrates. For sample preparation, 20 mL of hot deionized water (60–70 °C) was added to 20 g of homogenized pulp in a 50 mL volumetric flask with 5 mL of 0.1% sorbitol as the internal standard. After the mixture was shaken for 30 min, deionization water was added to the mark. Then an aliquot was centrifuged at 12300*g* for 10 min. Two milliliters of the filtered pulp was desalted by adding 1 g of a cationic resin (Amberlite IR-120, Fluka Chemie AG, Buchs, Switzerland) with 1 g of an anionic resin (Amberlite IR-96, Fluka). Samples were shaken occasionally during a 60 min desalting period. An aliquot (≈1 mL) of the solution was centrifuged at 11600*g* for 10 min, and 50 μL was injected into the chromatograph.

Organic acids (malic, citric, lactic, acetic, succinic, and formic acid) were analyzed by HPLC. The system was the same as mentioned above for the assessment of sugars. An Aminex HPX-87H [300 mm × 7.8 mm (inside diameter), Bio-Rad Laboratories] column, held at 65 °C, with 0.005 M H_2SO_4 as the mobile phase at a flow rate of 0.7 mL/min was used. The sample preparation was conducted in the same way as sugar determination, deleting the desalting step and without the additional sorbitol as the internal standard. Concentrations were calculated by comparing peak heights with those of external standards for each compound.

Statistical Analysis. Each olive sample (object) was considered an assembly of seven variables represented by fiber, glucose, fructose, mannitol, succinic acid, lactic acid, and acetic acid. Variables studied but not included in the analysis were sucrose and malic acid because they were always absent and citric acid because it was found in only a few samples. These variables formed a data vector which represented an olive sample. Data vectors belonging to the same group (elaboration style or cultivar) were analyzed. The group was termed a category. Differences within the elaboration styles between factories were not considered in the study and are included in the error term. The database from the analysis of these carbohydrate-related compounds was thus arranged in a 134 × 7 (cases × variables) matrix, since fiber data were twice the amount of 134, the average of two randomly selected duplicates which were used to construct a matrix with the same number of cases for all variables.

Original data were studied by multiple analyses of variance (MANOVA) to test overall differences between groups across variables.

Pattern recognition tools used in this work were as follows.

Autoscale. The variables were standardized (*15*) according to

$$y_{mj} = \frac{(x_{mj} - \bar{x}_m)}{s_m}$$

where y_{mj} is the value j for the variable m after scaling, x_{mj} is the value of the variable j before scaling, $\bar{x}_m$ is the mean of the variable, and s_m is the standard deviation for the variable. The result is a variable with zero mean and a unit standard deviation. The chemometric study was carried out using this standardized data.

Feature Selection. The selection of variables containing the most powerful information for the correct classification of olive commercial presentations in the three (types) or eight (cultivars) categories was carried out on the basis of a canonical analysis, using the backward stepwise analysis option. 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 and the minimum tolerance at 0.01, and no variable was forced to enter in any model.

Linear Discriminant Analysis (LDA). LDA is a supervised technique that provides a classification model characterized by a linear dependence of the classification scores with respect to the descriptors (groups previously defined). LDA assumes an *a priori* knowledge of the class membership of each sample in a training set. Two very distinct purposes and procedures for conducting discriminant analysis exist: discriminant predictive analysis (which involves only the derivation of the linear discriminant functions) and discriminant classification analysis (to evaluate the previous linear functions to classify current and future samples). To measure the classification power of the analytical data, the number of individuals correctly predicted to belong to the assigned group was calculated, considering that prior probabilities were proportional to the number of samples in each group. A leaving-one-out cross-validation procedure was performed to assess the performance of the classification rule (*16*).

Confusion Matrix Analysis. The computation of the confusion matrix has traditionally been the final step in the discriminant analysis. In this work, the confusion matrix, viewed as a contingency table, was also subject to further analysis, specifically with respect to the observed correct classification, by the conventional χ^2

$$\chi^2 = \sum_i \sum_j \frac{(o_{ij} - e_{ij})^2}{e_{ij}}$$

where o_{ij} is the observed number of samples classified in cell ij; $e_{ij} = (n_i.xn._j)/n$, where $n_i.$ is the number of samples classified in row i, $n._j$ the number of samples in column j, and n the total number of samples. As usual, the degree of freedom was $(i - 1)(j - 1)$.

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

RESULTS

Overall Contents of Fiber, Sugars, and Organic Acids in Table Olives. The overall distribution of dietary fiber, sugars, and organic acids is shown in **Figure 2**. Data from these variables always showed a skewed distribution toward the right (higher values).

Table olives have a moderate average content (2.75 ± 0.67 g/100 g e.p.) of dietary fiber (**Figure 2**). Most samples (45%) had between 2.0 and 2.5 g/100 g e.p.; 25% had between 2.5 and 3.0 g/100 g e.p, and the rest of the samples had higher contents.

Sucrose was absent in all commercial presentations, and the concentrations of the other sugars were also low. The average glucose content was 9.6 mg/100 g e.p. Most of the samples (72%) had between 0 and 11 mg/100 g e.p., while 26% had between 11 and 55 mg/100 g e.p. (**Figure 2**). Only one sample (Manzanilla stuffed with hot pepper) had a high proportion (99–110 mg/100 g e.p.) because of the use of partially fermented stuffing material. The average value of glucose was 14.88 mg/100 g e.p.; 75% of samples had less than 17.4 mg/100 g e.p., 21% between 17.4 and 69.7 mg/100 g e.p., and 3% between 139.5 and 174.3 mg/100 g e.p. The average mannitol concentration was 19.33 mg/100 g e.p.; most of the samples (78%) had less than 23.4 mg/100 g e.p., 19% between 23 and 106.8 mg/100 g e.p., and 2% between 169.3 and 211.0 mg/100 g e.p.

Malic acid was never found. Citric acid was found in only a few samples: Manzanilla pitted (100 mg/100 g e.p.) and stuffed with anchovy strips (150 mg/100 g e.p.), salmon strips (90 mg/100 g e.p.), garlic (165 mg/100 g e.p.), hot pepper (125 mg/100 g e.p.), ham paste (126 mg/100 g e.p.), and cheese (130 mg/100 g e.p.); Hojiblanca stuffed with red pepper strips (75 mg/100 g e.p.); and Aloreña seasoned in fresh (120 mg/100 g e.p.). Succinic acid was present in a limited number of samples and always at low concentrations (<40 mg/100 g e.p.); its average was 7.89 mg/100 g e.p. (**Figure 2**). Lactic acid (mean value of 230.08 mg/100 g e.p.) was widely found in olives; 22% of the samples had proportions between 0 and 70 mg/100 g e.p. (ripe olives) and 65% between 136.2 and 408.6 mg/100 g e.p. A few samples exhibited higher concentrations (676.7–681.0 mg/100 g e.p.). Acetic acid was found at concentrations between 0 and 102.5 mg/100 g e.p. in 62% of the commercial presentations, between 102.5 and 200.1 mg/100 g e.p. in 25%, and between 200 and 500 mg/100 g e.p. in 13%; only 1% of the samples exhibited high levels (883–1000 mg/100 g e.p.).

Effect of Processing Style. Processing, regardless of cultivars and presentations, had a significant ($p < 0.05$) effect on fiber, mannitol, and succinic, lactic, and acetic acid contents (**Table 1**). Overall, green olives had the lowest mean concentration of fiber (2.62 g/100 g e.p.), followed by ripe (2.90 g/100 g e.p.) and directly brined olives (3.33 g/100 g e.p.). Average glucose contents for the same styles were 8.89, 12.95, and 9.47 mg/100 g e.p., respectively, but differences were not significant. The fructose concentrations were 17.87, 10.97, and 4.08 mg/100 g e.p. for green, directly brined, and ripe olives, respectively. Mannitol was the sugar found in the greatest proportion with averages of 13.12 (green), 54.90 (directly brined), and 17.18 (ripe) mg/100 g e.p. The average content of succinic acid in green and directly brined olives was 8.80 and 11.78 g/100 g e.p., respectively, but succinic acid was not found in ripe olives. A great difference was observed between the concentrations of lactic acid in green (295.12 mg/100 g e.p.), directly brined (125.17 mg/100 g e.p.), and ripe olives (12.29 mg/100 g e.p.). The acetic acid content in green olives (mean, 119.85 mg/100 g e.p.) was lower than the lactic acid content, but acetic acid concentrations in directly brined (mean, 201.76 mg/100 g e.p.) or ripe olives (mean, 72.89 mg/100 g e.p.) were higher than lactic acid concentrations.

Effect of Cultivars within Processing Styles. Glucose and fructose were not detected in some commercial presentations. Mannitol was the only sugar (alcohol) consistently found in table olives (**Table 2**). Its content in the different cultivars ranged from 3.06 to 111.36 mg/100 g e.p.; its highest content was found in directly brined olives, especially Aloreña (111.36 mg/100 g e.p.) and Hojiblanca (88.63 mg/100 g e.p.) cultivars. The highest lactic acid concentrations were detected in green olives: Gordal

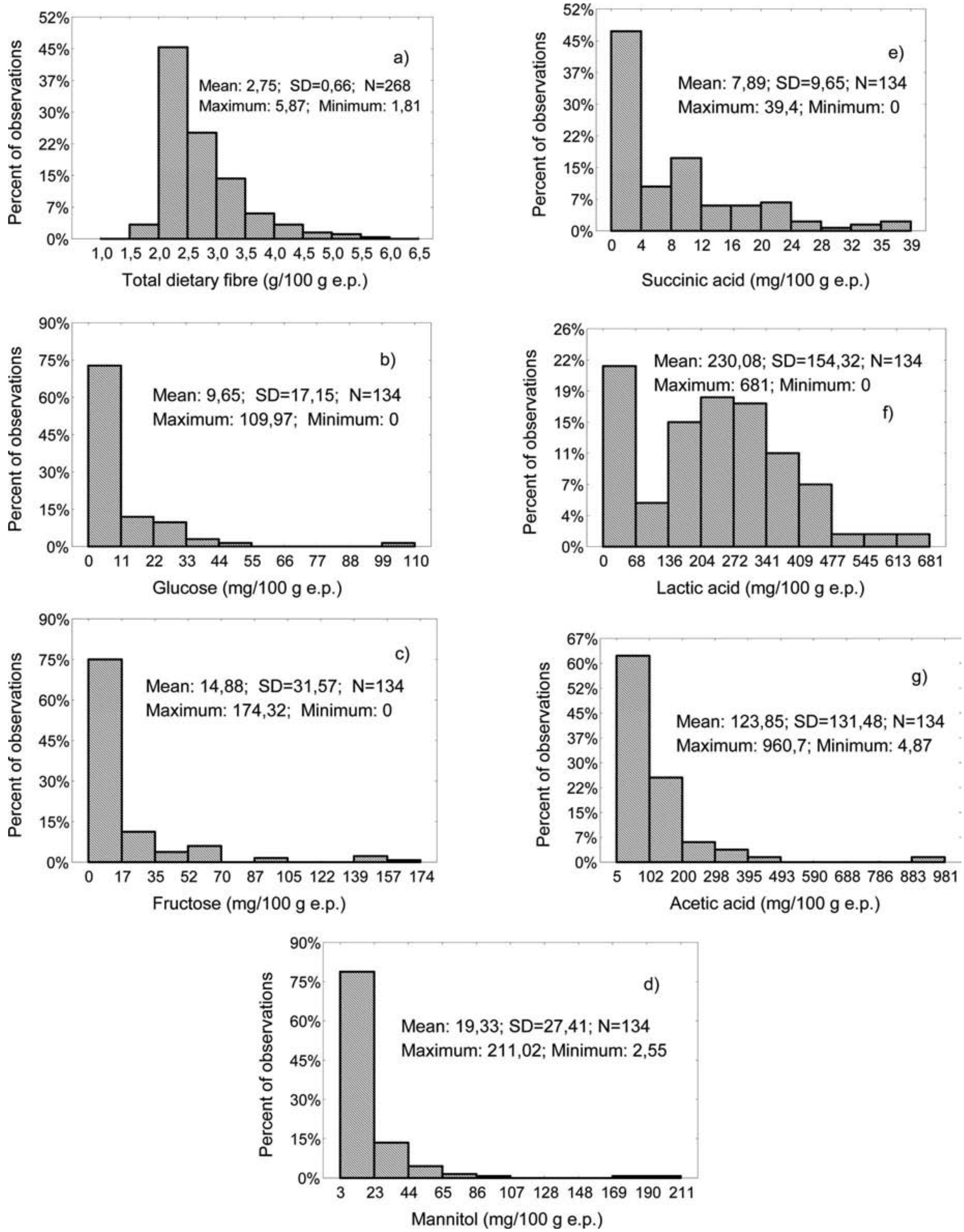

Figure 2. Distribution of (**a**) total fiber (g/100 g e.p.), (**b**) glucose (mg/100 g e.p.), (**c**) fructose (mg/100 g e.p.), (**d**) mannitol (mg/100 g e.p.), (**e**) succinic acid (mg/100 g e.p.), (**f**) lactic acid (mg/100 g e.p.), and (**g**) acetic acid (mg/100 g e.p.). Values were calculated from commercial presentations of Spanish table olives.

(409.05 mg/100 g e.p.), Hojiblanca (327.53 mg/100 g e.p.), Carrasqueña (296.72 mg/100 g e.p.), and Manzanilla (248.87 mg/100 g e.p.), which formed a statistically homogeneous group. Lactic acid was present in directly brined Manzanilla olives at the same level (289 mg/100 g e.p.) as in green olives but at lower concentrations than in the rest of the directly brined cultivars. Lactic acid was also found in cultivars processed as ripe olives (4.37–17.41 mg/100 g e.p.) but without significant differences among them. Acetic acid was always present in proportions in all cultivars regardless of the processing method. Its highest levels were observed in directly brined Verdial, Gordal, Arbequina, and Manzanilla cultivars. In addition, this acid was found, in combination with lactic acid, in green olives especially from Gordal and Carrasqueña cultivars. The acetic acid concentration in ripe olive cultivars ranged from 57.57 to 97.61 mg/100 g e.p.

Use of the Carbohydrate-Related Compounds for the Chemometric Evaluation of Table Olives. The multivariate

Table 1. Average Content of Fiber, Sugars, and Organic Acids, According to Processing Styles and Statistical Evaluation of Equity

compound	green	directly brined	ripe	difference: *F* value	difference: probability (*p*)
fiber[a]	2.62	3.33	2.90	10.38	<0.001
glucose[b]	8.99	9.47	12.95	0.44	0.665
fructose[b]	17.87	10.97	4.08	1.76	0.176
mannitol[b]	13.12	54.90	17.18	23.73	<0.001
succinic acid[b]	8.80	11.78	0.00	9.73	<0.001
lactic acid[b]	295.12	125.17	12.29	63.01	<0.001
acetic acid[b]	119.85	201.76	72.89	4.49	<0.001

[a] Expressed as grams per 100 g e.p. [b] Expressed as milligrams per 100 g e.p.

analysis of variance of the data vectors representing all olive samples (which did not include malic and formic acids, which were never detected, or sucrose and citric acid, found in only a limited number of commercial presentations) showed that there were significant differences among processing styles (**Table 2**) and cultivars (data not shown), except for glucose and fructose. Then the data were appropriate to be used in a chemometric analysis. Canonical and linear discriminant analyses were applied to the standardized matrix of data.

The most discriminant compounds for categorizing samples into elaboration styles, according to the canonical analysis, were fiber and mannitol as well as succinic, lactic, and acetic acids. Their contribution to discrimination can be estimated from the absolute coefficients of the canonical discriminant functions (standardized within variance). They were

$$\text{function } 1 = 0.67 \times \text{fiber} - 0.46 \times \text{glucose} + 0.52 \times \text{mannitol} + 0.33 \times \text{succinic} - 0.89 \times \text{lactic} + 0.36 \times \text{acetic}$$

$$\text{function } 2 = 0.22 \times \text{fiber} - 0.42 \times \text{glucose} + 0.67 \times \text{mannitol} + 0.33 \times \text{succinic} + 0.53 \times \text{lactic} + 0.39 \times \text{acetic}$$

Results indicate that the most discriminating compounds were as follows: fiber, glucose, and lactic acid in Function 1 and mannitol and acetic acid in Function 2. By applying these functions to the different olive samples, we calculated their corresponding scores for each function. A plot of the scores versus the canonical functions makes it possible to visualize their ability to discriminate among elaboration styles (**Figure 3**). Samples of green olives were characterized by low values of Function 1 and Function 2. Directly brined olives had higher scores for Function 1 than green and similar or slightly lower for Function 2. Ripe olive score values ranged from 0 to 2 units in Function 1 and from 1 to 2 units in Function 2; all samples from ripe olives were clearly separated from the rest, but a few green olive samples were situated within the region of directly brined olives. The structure matrix (**Table 3**), which represents the highest absolute correlation with each function, indicated that Function 1 could be related to fiber (0.306) and mannitol (0.410) while succinic (0.508), lactic (0.689), and acetic (0.332) acids had the highest correlation with Function 2 (acid-related function).

The confusion matrix associated with the discriminant analysis between styles showed good specificity and sensibility. The overall success of classification was 95.5% (**Table 4**). The most accurately characterized style was ripe olives with 100% specificity and 91% sensibility. Green olives were also well-classified, with high specificity (98%) and sensibility (98%). Directly brined olives had the lowest specificity (78%) and sensibility (88%). Cross validation showed similar results, so the deduced predictive classification functions were able to use the differences between carbohydrate-related compound concentrations among styles not only to distinguish between them but also to classify further unknown samples, although this study was focused mainly on the demonstration that the elaboration styles may introduce detectable differences in compositions among them.

For the classification of the samples into cultivars, the retained compounds were mannitol and succinic, lactic, and acetic acids. Therefore, four standardized canonical functions were determined:

$$\text{Function } 1 = 0.64 \times \text{fiber} + 0.501 \times \text{mannitol} + 0.34 \times \text{succinic} + 0.71 \times \text{lactic}$$

$$\text{Function } 2 = -0.13 \times \text{fiber} + 0.39 \times \text{mannitol} + 0.90 \times \text{succinic} + 0.18 \times \text{lactic}$$

$$\text{Function } 3 = -0.34 \times \text{fiber} + 0.78 \times \text{mannitol} - 0.53 \times \text{succinic} + 0.64 \times \text{lactic}$$

$$\text{Function } 4 = 0.75 \times \text{fiber} - 0.23 \times \text{mannitol} - 0.12 \times \text{succinic} + 0.54 \times \text{lactic}$$

The successive functions showed the largest absolute correlation (structure matrix) with mannitol, succinic, lactic, and fiber, respectively.

The retention of only four discriminating variables (and, as result, the deduction of the same number of canonical functions) indicates that differences between cultivars were limited. The two- or three-dimensional plottings of the sample scores versus the corresponding canonical functions did not lead to a net separation of cultivars. According to the confusion values (**Table 5**), 100% sensibility was obtained for Verdial and Arbequina cultivars while their specificities were 100 and 50%, respectively. Manzanilla obtained a good classification in sensibility (89.70%) but lower in specificity (61.00%). Aloreña had a sensibility of 50.00% and a slightly higher specificity (66.66%). Hojiblanca had very low sensibility and moderate specificity (44.00%). The worst results were obtained for Carrasqueña and Cacereña which were always misclassified (null sensibility and specificity). Results relative to cultivars with a small number of samples must be assessed with caution, but their commercial presentations on the market were reduced to the analyzed samples. Cross validation led to only slightly worse results (overall 60.4% correct classifications). Therefore, the prediction ability of the model to differentiate among cultivars was worse than that observed in the classification according to elaboration styles.

This is a case in which the evaluation of the confusion matrix is difficult. An analysis of results with respect to those expected by chance may be of interest. The observed overall correct classification was 61.20%. The calculation of the expected cases (e_{ij}) per cell and the overall χ^2 led to a value of 194.05 with $p < 0.001$ (49 degrees of freedom). Then, even in the case of classification by cultivars, the percentage of correct sample assignments was better than that which could have been obtained by chance.

DISCUSSION

The declaration of fiber and sugars in nutritional labeling is compulsory for most countries (*17*, *18*), and these compounds and organic acids must be considered to estimate the energy in the European Union food label (*18*).

The proportion of soluble fiber was always negligible with respect to the insoluble fraction, so the total fiber in table olives was, in practice, insoluble fiber. Fiber in table olives (average, 2.75 g/100 g e.p.) was comparable to that reported for other

Table 2. Average Content of Fiber, Sugars, and Organic Acids, According to Cultivars within Elaboration Styles (type III decomposition)[a]

style	cultivar	fiber[b]	glucose[c]	fructose[c]	mannitol[c]	succinic acid[c]	lactic acid[c]	acetic acid[c]	N[d]
green	Gordal	2.55 (0.06) a	11.39 (3.23)	41.01 (12.63)	17.66 (1.52) a	15.45 (2.08) ab	409.05 (25.75) b	216.33 (53.21) abc	22
green	Manzanilla	2.63 (0.07) a	10.08 (2.78)	13.55 (2.93)	11.33 (1.58) a	6.29 (0.82) ab	248.87 (11.48) b	88.60 (7.40) ab	60
green	Carrasqueña	2.37 (0.04) a	nd[e]	nd[e]	15.25 (5.51) a	3.94 (1.85) ab	296.72 (33.71) b	121.44 (10.37) ab	6
green	Hojiblanca	2.82 (0.11) a	1.00 (0.65)	nd[e]	12.42 (1.55) a	12.98 (2.38) ab	327.53 (30.01) b	87.71 (12.92) ab	8
brined	Gordal	2.30 (0.05) a	nd[e]	nd[e]	22.45 (0.68) a	18.82 (3.91) b	31.67 (1.20) a	355.20 (0.40) bc	2
brined	Manzanilla	2.94 (0.07) a	3.64 (1.16)	5.10 (1.80)	48.00 (6.07) a	12.31 (7.81) ab	289.33 (49.69) b	204 (30.96) abc	6
brined	Hojiblanca	2.92 (0.13) a	27.57 (1.82)	62.07 (3.53)	88.63 (6.94) b	nd[e]	nd[e]	86.54 (1.26) ab	2
brined	Arbequina	4.06 (0.16) b	6.39 (0.01)	nd[e]	10.02 (0.01) a	nd[e]	56.73 (0.01) a	275.10 (0.01) abc	2
brined	Aloreña	4.20 (0.15) b	20.19 (6.06)	10.67 (6.18)	111.36 (51.00) b	5.41 (3.24) ab	31.56 (18.64) a	34.28 (2.78) a	4
brined	Verdial	2.97 (0.04) a	nd[e]	nd[e]	6.27 (0.01) a	39.40 (0.01) c	107.00 (0.01) a	416.30 (0.01) c	2
ripe	Gordal	2.28 (0.09) a	nd[e]	nd[e]	3.06 (0.01) a	nd[e]	4.37 (0.19) a	91.91 (2.36) a	2
ripe	Manzanilla	3.28 (0.10) a	7.13 (0.10)	nd[e]	5.39 (2.03) a	nd[e]	16.72 (0.34) a	97.61 (4.30) a	2
ripe	Carrasqueña	2.66 (0.11) a	23.76 (6.41)	10.31 (5.95)	15.73 (0.88) a	nd[e]	12.27 (2.09) a	68.50 (11.20) ab	4
ripe	Hojiblanca	3.26 (0.06) a	16.30 (5.33)	3.20 (1.02)	28.57 (6.18) a	nd[e]	8.37 (2.89) a	76.56 (25.83) ab	6
ripe	Cacereña	2.98 (0.06) a	8.66 (5.48)	3.51 (2.22)	15.38 (4.14) a	nd[e]	17.41 (0.64) a	57.57 (15.86) a	6

[a] Values are means, with the standard error in parentheses. Values followed by the same letter(s) constitute homogeneous groups at $p < 0.05$. [b] Expressed as grams per 100 g e.p. [c] Expressed as milligrams per 100 g e.p. [d] Number of samples for sugars and organic acids (number of samples for fiber was twice these figures); differences between cultivars within styles were always significant at $p < 0.05$ except for glucose. [e] Not detected.

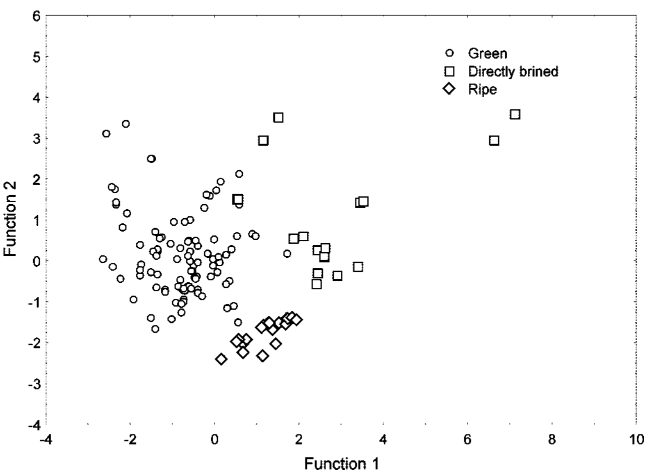

Figure 3. Plot of Spanish table olive commercial presentation sample scores as a function of the two canonical discriminant functions, according to elaboration styles.

Table 3. Structure Matrix of the Canonical Discriminant Study among Styles

parameter	Function 1	Function 2
fiber	0.306[b]	0.112
glucose[a]	–	–
fructose[a]	–	–
mannitol	0.410[b]	0.403
succinic acid	−0.050	0.508[b]
lactic acid	−0.657	0.689[b]
acetic acid	0.095	0.332[b]

[a] Variables not in the model. [b] Largest absolute correlation between each variable and any discriminate function.

Table 4. Classification Results[a]

actual group	predicted group membership 1	2	3	total	% correct (sensibility)
1	94 (92)	2 (4)	0 (0)	96	98 (96)
2	2 (3)	14 (12)	2 (3)	18	78 (67)
3	0 (0)	0 (0)	20 (20)	20	100 (100)
total	96 (95)	16 (16)	22 (23)	134	96 (93)
% correct (specificity)	98 (97)	88 (75)	91 (87)		

[a] Elaboration styles were coded as 1 (green Spanish style), 2 (directly brined), and 3 (ripe olives). Results from cross validation are given in parentheses; 95.5% of original group cases are correctly classified, and 92.5% of cross-validated grouped cases are correctly classified.

vegetables or fruits (*9*). The fiber content in table olives is exceeded only by that of dried fruits [figs, 9.0; apricots, 8.0; raisins, 70.0; prunes, 5.0, (in grams per 100 g e.p.)]; however, the contents of these products can be similar to those of dried or dehydrated olives.

The average total sugar concentration in table olives was fairly low because these compounds are consumed by microorganisms or eliminated in the diverse lye and washing treatments during processing. The highest concentrations of mannitol were related to directly brined olives.

The organic acids present in fresh olives were practically removed during processing (*2*), and succinic acid was the only acid from the raw material observed in green and directly brined olives (<40 mg/100 g e.p.). Therefore, acids found in the final products were those produced during fermentation or storage or added to control the pH (*2*). Apparently, citric acid, authorized to control the pH and prevent oxidation (*19*) in table olives, had a limited use because it was detected in only a reduced number of samples.

Processing had a significant effect (**Table 1**) on all carbohydrate-related compounds, except glucose and fructose. Jiménez et al. (*20*) reported significant differences between fresh and processed fruits. Fiber was significantly lower in green and ripe olives, possibly due to the effect of the lye treatment. Marsilio et al. (*21*) observed that lye removed the epicuticuler waxes of the cuticle in green olives and reduced its thickness and the level of cellular cohesion. Jiménez at al. (*22*) found that lye affected the pectin fractions, the water-soluble polysaccharides (which almost disappeared), decreased the degree of esterification in oxalate-soluble rhamnogalacturonans, and reduced the molecular weights of the main polysaccharide components of the hemicelluloses.

Chung et al. (*23*) studied the changes in the cell wall of ripe olives and related the differences in texture to changes in the total pectin and protopectin contents, which were leached out during curing and retorting.

Dietary fiber in ripe olives was higher than in green olives in spite of the more numerous lye and washing treatments used during its processing. This behavior may be due to the presence of higher levels of Ca^{2+} in ripe olives because of the use of this cation in the previous storage solution. Polymerization of the phenolics in the olive pulp during darkening may also prevent the solubilization of the glycosidic polymers and produce a complex group of substances with a wide range of

Table 5. Confusion Matrix of the Discriminant Analysis of the Different Cultivars According to Carbohydrate-Related Compound Concentrations[a]

		predicted cultivar									
actual cultivar	$n_{i\cdot}$	G	M	Cr	H	Ar	Al	Vr	Cc	sensibility (%)	$p = n_{i\cdot}/n$
G	26	11 (11)	15 (14)	0 (0)	0 (1)	0 (0)	0 (0)	0 (0)	0 (0)	42.30	0.1940
M	68	3 (2)	61 (61)	0 (0)	2 (2)	2 (2)	0 (0)	0 (1)	0 (0)	89.70	0.5075
Cr	10	0 (0)	10 (10)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0.00	0.0746
H	16	1 (1)	9 (9)	0 (0)	4 (3)	0 (0)	1 (1)	0 (0)	1 (2)	25.00	0.1194
Ar	2	0 (0)	0 (0)	0 (0)	0 (0)	2 (2)	0 (0)	0 (0)	0 (0)	100.00	0.0149
Al	4	0 (0)	0 (0)	0 (0)	2 (2)	0 (0)	2 (2)	0 (0)	0 (0)	50.00	0.0298
Vr	2	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	2 (2)	0 (0)	100.00	0.0149
Cc	6	0 (0)	5 (5)	0 (0)	1 (1)	0 (0)	0 (0)	0 (0)	0 (0)	0.00	0.0448
specificity (%)		73.33	61.00	0.00	44.44	50.00	66.66	100.00	0.00	61.20	
$n_{\cdot j}$		15	100	0	9	4	3	2	1	$n = 134$	
$\alpha = n_{\cdot j}/n$		0.1119	0.7463	0.0000	0.0672	0.0299	0.0223	0.0149	0.0075		

[a] Classifications obtained from cross validation are given in parentheses. Cultivars are coded as follows: G, Gordal; M, Manzanilla; Cr, Carrasqueña; H, Hojiblanca; Ar, Arbequina; Al, Aloreña; Vr, Verdial, and Cc, Cacereña. Overall correct classification, 61.20%; overall correct classification after cross validation, 60.40%. Overall $\chi^2 = 195$ with 49 df, (8 – 1)(8 – 1) ($p < 0.001$).

molecular masses associated with protein and dietary fiber (*24*). Fiber content in directly brined olives was high; however, these olives may suffer texture deterioration when incised or when packed in an acidic pH. The attack of some microorganisms or enzymes from natural ingredients of the seasoning materials may also facilitate cell wall losses (*25*).

Processing styles affected sugar contents. The sugar concentration in green olives was fairly low (due to its exhaustion during fermentation). Directly brined olives exhibited the highest sugar content because of their partial fermentation and the more difficult solubilization of this compound (absence of lye treatment). The numerous lye and washing treatments used in ripe olives are responsible for the low sugar content found in samples from this style. The high levels of mannitol observed in all commercial presentations mean a limited use of this compound by the microorganisms in the previous fermentation or storage processes; its lowest content in green and ripe olives can be due to its higher degree of removal by the lye and washing treatments applied in these styles.

Apparently, the levels of succinic acid were inversely proportional to treatment strength, and it was completely removed from ripe but not from green or directly brined olives. Green olives retained the highest content of lactic acid (due to its production during fermentation), followed by directly brined products. The presence of lactic acid in ripe olives may be due to a residue from the previous storage phase or its use for a final pH adjustment. Acetic acid was widely applied in directly brined olives, because its flavor resembles traditional homemade seasoned olives, but its use in green olives was reduced to products in which cucumbers or other pickles were also present as stuffing materials or ingredients. Acetic acid is also more frequently used in ripe olives than lactic acid. The sum of all acid contents was in agreement with the titratable acidity reported in a recent survey for green (0.42 and 0.75 g of lactic acid/100 mL of brine) and seasoned olives (0.27 – 0.85 g of lactic/100 mL of brine) (*26*).

Apparently, green olive processing produces an approximation of the fiber content by applying more severe treatments to those cultivars with higher cell wall materials (*2*). In fact, dietary fiber content in Gordal, Manzanilla, and Hojiblanca was similar, but the lye strength used in Carrasqueña must be excessive because its fiber content was significantly lower than those in Hojiblanca and Manzanilla.

All directly brined cultivars retained their consistency well because of the absence of a lye treatment in their elaboration, and Arbequina and Aloreña had the highest values of fiber content. However, Manzanilla and Hojiblanca prepared according to this style exhibited values only slightly above those observed when they were prepared as green olives.

Cultivar determines the ripe olive processing treatments, which are designed with the purpose of producing homogeneous final products. The objective was not completely achieved in the case of Hojiblanca cultivar which had a higher total fiber content than Manzanilla, Cacereña, Carrasqueña, and Gordal cultivars. For the same cultivars, ripe olives usually have slightly higher (but not significant) fiber content than green olives because the added divalent cations in its previous storage phase fixed the cell wall material through calcium bridges and prevented softening (*22*, *24*).

Green Carrasqueña and Hojiblanca olives are usually subjected to intense washings before packing. As a result, glucose and fructose were not found in samples from this style, but the same sugars were present in green Gordal and Manzanilla cultivars, especially in commercial presentations stuffed with partially fermented ingredients. In directly brined olives and ripe olives, the diverse contents of glucose and fructose, according to cultivar, reflected heterogeneous industrial practices (**Table 2**). Mannitol was present at similar levels in all green, all ripe, and some directly brined olive cultivars, but the level was high in Aloreña (packed in fresh) (**Table 2**). The cultivar, within the same elaboration process, also had a significant effect on the succinic acid content in directly brined olives (the highest value corresponded to Verdial) but not in green samples (which exhibited similar concentrations) or ripe olives (in which it was absent). There was no effect from cultivars on lactic and acetic acid levels. The values of these acids in green Manzanilla olives correlate well with those of titratable acidity reported by Sánchez et al. (*27*).

Most of the green olive commercial presentations within cultivars had the same fiber contents because of a reduced proportion of stuffing material (6%, w/w, with respect to the edible portion) and a similar proportion of fiber in them. However, some materials may contribute to an increase or decrease in fiber content in specialties such as Gordal and Manzanilla olives stuffed with almonds (≈3.5 g/100 g e.p.), hazelnuts (≈3.5 g/100 g e.p.), hot red peppers (≈4.0 g/100 g e.p.), and a blend of olives with cappers (5.0 g/100 g e.p.). In general, fiber values found in this work for green olives were considerably higher than those reported by Vázquez Ladrón et al. (*28*) for the same product packed at laboratory scale. Some specific commercial presentations may have noticeable sugar levels like green Gordal and Manzanilla cultivars stuffed with natural hot pepper (which showed the highest sugar concentration) and green Gordal cultivar stuffed with garlic.

The effect of commercial presentations on carbohydrate-related compounds in directly brined olives was reduced due to the limited number of samples in this style. Fresh seasoned Aloreña olives had the highest total sugar content (169.3–211.0 mg/100 g e.p.). Fiber contents found in this work for directly brined olives were 50% higher than those reported by Nosti Vega et al. (*29*) and higher than those included in the Food Composition and Nutrition Tables (*9*) for green marinated olives.

Commercial presentations influenced the type and proportions of acid used. Lactic acid was the acid used in most green olive presentations. The use of a high proportion of acetic acid was characteristic of green stuffed with whole cucumber Gordal cultivar and directly brined Gordal cultivar. The highest proportion of acetic acid was related to seasoned Verdial cultivar. In ripe olives, the highest acetic acid content was observed in whole Hojiblanca olives.

The selection of the minimum number of variables to reach a correct classification according to styles was achieved by choosing the features which contain the most discriminant information for the classification. The classification model distinguished among elaboration styles fairly well. However, the classification according to cultivar, although significantly better than by chance, as demonstrated by the χ^2 test, led to some misclassification. Most of the Manzanilla samples were correctly classified, but many samples from Gordal, Carrasqueña, and Hojiblanca were misclassified as Manzanilla. In directly brined olives, cultivars exclusively devoted to this style (Arbequina, Aloreña, and Verdial) were correctly classified but Cacereña, only used to prepare ripe olives, was mainly misclassified as Manzanilla. This may indicate that the variables that were studied were more influenced by elaboration styles than by cultivars.

This is the first time that total dietary fiber, sugar, and organic acid contents and the influence of processing styles, cultivars, and commercial presentations on their contents in table olives have been studied. Table olives contain a moderately high percentage of total dietary fiber with an apparently high digestibility, comparable to those values found in other similar fruits and higher than those of many green vegetables. In particular, green olives and directly brined olives are rich in polyphenols, so they could be included in the recently defined antioxidant dietary fiber products (*30*), which are characterized by the combined high content of both fiber and polyphenols. Glucose and fructose levels were negligible in most commercial presentations, and only mannitol was always found in noticeable concentrations. Therefore, table olives can be considered low-sugar or even sugar-free products. The presence of organic acids was restricted to succinic (almost exclusively in green olives), lactic, and acetic acids. Lactic acid was mainly used in green olives, followed by directly brined olives and ripe olives (lowest proportion). Acetic acid was the most widely used organic acid in olives regardless of processing style and cultivar and was specific to green Gordal stuffed with cucumber. The presence of organic acids must then be considered when estimating the energetic value of table olives in those cases required by legislation.

ACKNOWLEDGMENT

We thank Amparo Cortés and Francisco Rodriguez for technical assistance.

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Received for review July 10, 2007. Revised manuscript received September 26, 2007. Accepted September 27, 2007. This work was supported by the Spanish Government (AGL-2003-00779 and AGL-2006-03540/ALI), the European Union (TDC-Olive), and Junta de Andalucía (through financial support to group AGR-125).

JF0720638