



The effects of cultivar and harvest year on the fatty alcohol composition of olive oils from Southwest Calabria (Italy)

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SUMMARY: The fatty alcohol (FAL) composition of olive oils produced in Southwest Calabria (Southern Italy) was analyzed over three harvest years 2005–2006–2007. Three autochthonous cultivars: Cassanese, Ottobratica, Sinopolese and seven allochthonous cultivars: Coratina, Itrana, Leccino, Nocellara Messinese, Nociara, Pendolino and Picholine were considered. Hexacosanol was always the main FAL occurring in the olive oil. The autochthonous cultivars were among those with the highest total FAL contents. Nevertheless, both autochthonous and allochthonous cultivars produced olive oil within the limit indicated by EU and IOC regulations. Cultivar affected the FAL content highly significantly or very highly significantly, whereas in a large part of the cases the harvest year did not influence the FAL content. Cultivar x harvest year influenced highly significantly or very highly significantly only C_{22-OH} , C_{23-OH} , C_{24-OH} , C_{25-OH} , odd chain FALs and the ratio even chain/odd chain FALs.

KEYWORDS: *Fatty alcohols; Minor components; Olive oil; Policosanols; Unsaponifiable*

RESUMEN: *Efectos de la variedad y año de cosecha sobre la composición de alcoholes grasos de aceites de oliva del Suroeste de Calabria (Italia).* Se ha analizado, durante tres cosechas 2005–2006–2007, la composición de alcoholes grasos (FAL) de aceites de oliva producidos en el suroeste de Calabria (sur de Italia). Se consideraron tres cultivares autóctonos: Cassanese, Ottobratica y Sinopolese y siete alóctonos: Coratina, Itrana, Leccino, Nocellara Messinese, Nociara, Pendolino y Picholine. Hexacosanol fue siempre el principal FAL en los aceites de oliva. Los cultivares autóctonos se encuentran entre los de mayor contenido total de FAL. No obstante, ambos cultivares, autóctonos y alóctonos producen aceites de oliva dentro de los límites de los reglamentos de la UE y del COI. Los cultivares afectan de manera altamente significativa o muy significativa al contenido de FAL, mientras que el año de cosecha no influyó en el contenido de FAL. Cultivar x año de cosecha influye de manera altamente significativa o muy significativa sobre C_{22-OH} , C_{23-OH} , C_{24-OH} , C_{25-OH} , al total de alcoholes grasos de cadena impar y a la relación: FALs cadena par/FALcadena impar.

PALABRAS CLAVE: *Aceite de oliva; Alcoholes grasos; Componentes menores; Insaponificable; Policosanoles*

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1. INTRODUCTION

Olive oil is mainly composed of glyceridic components (Giuffrè, 2013a) and minor components such as sterols (Giuffrè, 2012; Giuffrè *et al.*, 2012; Giuffrè and Louadj, 2013), waxes (Giuffrè, 2013b), polyphenols and tocopherols, which are generally used to characterize a mono-cultivar olive oil and to evaluate its chemical quality. Fatty alcohols (FALs) are contained in the minor component fraction and in particular in the unsaponifiable fraction. FALs are a useful parameter for classifying different categories of olive oil: oils with a wax content of between 300 mg·kg⁻¹ and 350 mg·kg⁻¹ are considered to be lampante olive oil if the total aliphatic alcohol content is less than or equal to 350 mg·kg⁻¹; oils with a wax content of between 300 mg kg⁻¹ and 350 mg·kg⁻¹ are considered to be crude olive-pomace oil if the total aliphatic alcohol content is above 350 mg·kg⁻¹ and if the erythrodil and uvaol content is greater than 3.5 % (EU, 2011; IOC, 2012). Grob *et al.* (1990) found a fatty alcohol content in raw solvent-extracted olive oils (725 mg·kg⁻¹) ten times higher than in extra virgin olive oil (73 and 61 mg·kg⁻¹). All FALs otherwise known as aliphatic alcohols in olive oil are a mixture of long chain fatty alcohols known as policosanols. The majority of these are even chain fatty alcohols (ECFALs): C_{22-OH}, C_{24-OH}, C_{26-OH}, and C_{28-OH}. Odd chain fatty alcohols (OCFALs) are present in minor amounts: C_{23-OH}, C_{25-OH}, and C_{27-OH}.

It is worth noting that the FAL composition changes in olive oil during ripe table olive processing with a decrease in docosanol and a substantial increase in octacosanol (López-López *et al.*, 2009). Octacosanol is the most common in olive oil extracted from commercial table olives of different cultivars processed by Spanish / Sevillian style, or untreated and directly brined with a limited fermentation, or by California style (López-López *et al.*, 2008). Octacosanol is very effective in lowering LDL and increasing HDL (Taylor *et al.*, 2003). In addition, it has been shown that policosanols are as effective as aspirin in terms of its antiaggregatory effects. Octacosanol also offers cytoprotective effects. This affords an opportunity for octacosanol to be taken as an alternative to aspirin in patients who have a history of or suffer from gastric irritation (Taylor *et al.*, 2003).

The aim of this paper was to study the fatty alcohol composition (docosanol C_{22-OH} - tricosanol C_{23-OH} - tetracosanol C_{24-OH} - pentacosanol C_{25-OH} - hexacosanol C_{26-OH} - heptacosanol C_{27-OH} - octacosanol C_{28-OH} - ECFALs - OCFALs - total FALs) of pressed olive oil from autochthonous and allochthonous olive cultivars growing in Southwest Calabria (Southern Italy), on the basis of the European (EU, 2011) and the I.O.C. (IOC, 2012) regulations. Particular emphasis was placed on the effect of cultivar and harvest year on fatty alcohol composition.

To our knowledge this is the first report to show the effects of cultivar and harvest year on fatty alcohol composition of olive oils from cultivars grown in Southwest Calabria (Southern Italy).

2. MATERIALS AND METHODS

2.1. Plant material and extraction procedure

The experiments were conducted over three harvest years 2005, 2006 and 2007 with the Cassanese, Coratina, Itrana, Leccino, Nociara, Ottobratica, Pendolino, Picholine and Sinopolese cultivars. The Nocellara Messinese cultivar was studied only in the 2005 harvest year. Cassanese, Ottobratica and Sinopolese are commonly cultivated in the Calabria Region (Southern Italy) for oil extraction. The other examined cultivars are allochthonous for this region. Olive trees were well managed and had no nutrient deficiency or pest damage. Fifteen 25–40 year old trees per cultivar were selected and labeled in mono-cultivar groves, situated in the area of Rizziconi (Southwest Calabria). This area, at an altitude of 100 m above sea level, is characterized by damp and rainy winters and hot summers. Each mono-cultivar grove was at least 3 km from the others. Olive sampling was conducted at biweekly intervals from October, when the fruit was 20% ripe, until fruit was no longer found on the trees. Freshly and manually harvested drupes (40 kg approximately per cultivar, 2.5 kg approximately per tree) were placed in a plastic container and immediately transported to the laboratory where they were cleaned to eliminate twigs and leaves and were washed in fresh water to remove dust. At this point, the olives were immediately processed in a laboratory mill “Mini 30” (AGRIMEC Valpesana, Calzaiolo, S. Casciano VP, Florence), with a capacity of 40 kg. First the olives were crushed with a hammer-mill. The resulting paste was mixed at a temperature between 15 and 20 °C for 35 minutes, then placed between a pile of circular metallic grids and pressed using a hydraulic press with a mild and continuous increase in pressure up to 200 bars. The liquid phase was submitted to separation by centrifugation and the obtained oil was filtered through filter paper. The oil was kept in 100 mL amber glass bottles and maintained in dark conditions at 15–20 °C, until analysis.

2.2. Chemicals

Standard samples of 1-docosanol (behenyl alcohol), 1-tricosanol (tricosyl alcohol), 1-tetracosanol (lignoceryl alcohol), 1-heptacosanol, 1-octacosanol (octacosoyl alcohol) from Sigma-Aldrich (St. Louis, MO (USA)), and 1-pentacosanol (pentacosoyl alcohol) from GmbH (Germany), and 1-hexacosanol (ceryl alcohol) from J&K scientific Ltd. (China) were used as references. TLC silica gel plates were

from Merck S.p.A., (Milan, Italy). All other reagents were from Carlo Erba, (Milan, Italy).

2.3. Methods

2.3.1. Determination of fatty alcohols

FAL composition was determined as described in Annex XIX of the Consolidated Text on the characteristics of olive oil and olive-residue oil and on the relevant methods of analysis CONSLEG 2003 (CONSLEG, 2003). Olive oil (5g) was saponified with a 2 M ethanolic potassium hydroxide solution, using eicosanol (C_{20-OH}) as an internal standard; after boiling, 50 mL of distilled water was added. The reaction mixture was extracted three times with ethyl ether. The three ether extracts were introduced into a separating funnel and washed with distilled water (50 mL each time) until reaching a neutral reaction. The organic extracts were dried with anhydrous sodium sulphate and filtered. These extracts were evaporated to dryness using a rotary evaporator. The remaining residue was dissolved in 2 mL of chloroform, and then the FAL fraction was separated by TLC using a plate-developing chamber which contained hexane/diethyl ether 60:40 (v/v). After TLC separation, the silica plate was sprayed lightly and uniformly with 2,7-dichlorofluorescein. The FAL fraction was separated from the unsaponifiable extract by chromatography on a basic silica gel plate. The FALs recovered from the silica gel were transformed into trimethyl silyl ethers and analyzed by a gas chromatograph Perkin Elmer, Model 8600. The working conditions were: carrier gas (helium) 10 psi of pressure, auxiliary gas (hydrogen at 15 psi and air at 22 psi), split/splitless injector (operating in the split mode) temperature (280 °C), flame ionization detector (F.I.D.) temperature (290 °C), a capillary column SE 54 (30 m length \times 0.32 mm ID, 0.5 μ m film thickness, Mega, Milan - Italy) and an injection volume of 1 μ L. The temperature program used for the analysis was as follows: initial temperature as set at 180 °C, held for 2 min, and ramped at 2 °C \cdot min⁻¹ to 260 °C held for 15 min and ramped at 6 °C \cdot min⁻¹ to 270 °C. The identification of the compounds was based on a comparison of retention indices with those of standard samples and with literature data.

2.3.2. Statistical analysis

SPSS 15.0 software was used to determine the significant differences for all parameters. Two effects were taken into consideration, the cultivar and the harvest year. Data were analyzed by one-way and two-way analyses of variance (ANOVA) at 5% significance level and regression analysis. The Duncan test was used to determine the differences between cultivars. Bar graphs were constructed with Excel 2003.

3. RESULTS AND DISCUSSION

3.1. Fatty alcohol composition

In Tables 1 and 2 the different contents of FALs found in the ten studied cultivars are reported. The highest value of C_{22-OH} was found in Ottobratica (21.47 \pm 11.61 mg \cdot kg⁻¹) accounting for 15.51% of the total FALs, followed by Pendolino and Sinopolese (with 13.93 \pm 0.52 mg \cdot kg⁻¹ and 12.86 \pm 7.56 mg \cdot kg⁻¹, respectively), Coratina represented the lowest content with 4.40 mg \cdot kg⁻¹. The C_{22-OH} varied from 6.32% (Coratina) to 16.04% (Nocellara Messinese) in all the studied cultivars. The C_{23-OH} content was the highest in Sinopolese (3.33 \pm 3.06 mg \cdot kg⁻¹) and the lowest in Itrana (0.87 \pm 0.35 mg \cdot kg⁻¹). C_{23-OH} accounted for 0.93%-3.01% of the total FALs in all the studied cultivars. The C_{24-OH} was lowest in Nocellara Messinese 6.71 \pm 1.58 mg \cdot kg⁻¹ and highest in Pendolino (36.00 \pm 8.54 mg \cdot kg⁻¹), 5.36 times more and ranged from 14.84% of Coratina, to 31.13% of Pendolino. The greatest content of C_{25-OH} was observed in Cassanese with 8.80 \pm 5.00 mg \cdot kg⁻¹ (10.13% of the total FALs), the lowest content was detected in Nocellara Messinese with 1.56 \pm 0.58 mg \cdot kg⁻¹ (4.63% of the total FALs).

C_{26-OH} always made up the highest percentage and gave the highest quantity of all fatty alcohols in the cultivars studied. In particular, the highest content was found in Sinopolese (47.80 \pm 20.68 mg \cdot kg⁻¹) and the lowest content was found in Nocellara Messinese (12.75 \pm 11.19 mg \cdot kg⁻¹). By considering the results as a percentage, C_{26-OH} was the highest in Coratina (46.08%) and the lowest in Ottobratica (27.34%). It was observed that C_{27-OH} was always lower than 3.33 mg \cdot kg⁻¹ in Ottobratica and accounted for less of 3.93% in Nociara.

C_{28-OH} ranged between a minimum of 10.03 \pm 9.78 mg \cdot kg⁻¹ in Nocellara Messinese and a maximum of 30.53 \pm 13.42 mg \cdot kg⁻¹ in Ottobratica (three times more).

In all cases, ECFALs were in higher amounts than OCFALs with the maximum ratio in Pendolino (15.24 \pm 2.46) and in Leccino (13.19 \pm 8.87) and the minimum ratio in Cassanese (5.43 \pm 2.02). The greatest amounts of ECFALs were found in Ottobratica (119.94 \pm 43.10 mg \cdot kg⁻¹, 91.61%), in Pendolino (116.93 \pm 66.16 mg \cdot kg⁻¹, 93.61%) and in Sinopolese (112.40 \pm 30.44 mg \cdot kg⁻¹, 90.82%). OCFALs were found in the highest amount in Cassanese (13.86 \pm 5.63 mg \cdot kg⁻¹, 16.07%) and in the lowest quantity in Nocellara Messinese (3.44 \pm 1.56 mg \cdot kg⁻¹, 9.93%). Total FALs varied to a large extent according to cultivars, showing the highest amounts in Ottobratica (130.40 \pm 45.16 mg \cdot kg⁻¹), in Pendolino (124.60 \pm 70.28 mg \cdot kg⁻¹), in Sinopolese (123.73 \pm 32.49 mg \cdot kg⁻¹) and the lowest amount in

TABLE 1. The composition in fatty alcohols of the different cultivars [$\text{mg}\cdot\text{kg}^{-1}$]. The values represent the mean of five measurements \pm standard deviation. Rows followed by the same letter were not significantly different according to the multiple range Duncan test at $p \leq 0.05$

	Cassanese	Coratina	Itrana	Leccino	Nocellara Messinese	Nociara	Ottobratica	Pendolino	Picholine	Sinopolese
C _{22-OH}	9.00 \pm 5.30cde	4.40 \pm 1.92e	6.36 \pm 3.26de	10.87 \pm 5.66bed	4.81 \pm 1.54e	4.45 \pm 2.18e	21.47 \pm 11.61a	13.93 \pm 8.52b	5.73 \pm 2.02e	12.86 \pm 7.56bc
C _{23-OH}	1.94 \pm 0.80b	0.93 \pm 0.26c	0.87 \pm 0.35c	1.40 \pm 0.64bc	0.89 \pm 0.43c	1.53 \pm 1.18c	2.93 \pm 1.22a	0.93 \pm 1.03bc	1.13 \pm 0.92bc	3.33 \pm 3.06a
C _{24-OH}	23.40 \pm 9.76b	11.20 \pm 2.57c	10.80 \pm 2.34c	21.33 \pm 7.04b	6.71 \pm 1.58c	8.53 \pm 1.42c	33.43 \pm 18.38a	36.00 \pm 18.54a	10.87 \pm 3.40c	25.13 \pm 10.09b
C _{25-OH}	8.80 \pm 5.00a	3.40 \pm 1.72bc	2.00 \pm 0.76cd	2.94 \pm 1.36bcd	1.56 \pm 0.58d	2.07 \pm 0.88cd	4.00 \pm 0.93b	4.53 \pm 2.48b	1.87 \pm 0.74cd	5.00 \pm 1.41b
C _{26-OH}	30.60 \pm 15.90b	39.27 \pm 22.72ab	14.04 \pm 9.03c	33.58 \pm 16.73ab	12.75 \pm 11.19c	15.85 \pm 11.02c	34.51 \pm 14.35ab	45.03 \pm 28.98a	16.99 \pm 9.94c	47.80 \pm 20.68a
C _{27-OH}	3.12 \pm 2.21a	2.13 \pm 1.55abc	1.00 \pm 0.53cd	1.93 \pm 1.16abcd	0.99 \pm 0.94cd	2.00 \pm 1.60bcd	3.33 \pm 1.88a	2.20 \pm 1.66abc	0.93 \pm 0.46d	3.00 \pm 1.51ab
C _{28-OH}	12.21 \pm 7.23c	21.86 \pm 14.08ab	10.60 \pm 5.34c	17.07 \pm 10.60bc	10.03 \pm 9.78c	14.44 \pm 10.41c	30.53 \pm 13.42a	21.96 \pm 14.49b	11.27 \pm 5.23c	25.13 \pm 11.15ab
Totals	89.07 \pm 34.29b	83.33 \pm 39.85b	45.67 \pm 17.88c	89.13 \pm 28.97b	37.77 \pm 23.36c	48.27 \pm 21.37c	130.40 \pm 45.16a	124.60 \pm 70.28a	48.80 \pm 16.39c	123.73 \pm 32.49a
Σ ECFALs	75.21 \pm 30.14c	76.86 \pm 38.52c	41.73 \pm 16.70d	82.85 \pm 27.25bc	34.33 \pm 21.94d	43.27 \pm 19.51d	119.94 \pm 43.10a	116.93 \pm 66.16a	44.87 \pm 14.99d	112.40 \pm 30.44b
Σ OCFALs	13.86 \pm 5.63a	6.47 \pm 2.56cd	3.93 \pm 1.62de	6.28 \pm 2.51cd	3.44 \pm 1.56e	4.99 \pm 2.20de	10.46 \pm 3.12b	7.67 \pm 4.36c	3.93 \pm 1.67de	11.33 \pm 4.74b
ECFALs/ OCFALs	5.43 \pm 2.02d	11.88 \pm 6.73a	10.62 \pm 6.49ab	13.19 \pm 8.87a	9.98 \pm 2.52bc	8.67 \pm 2.28cd	11.47 \pm 2.79abc	15.24 \pm 2.46a	11.42 \pm 2.66abc	9.92 \pm 6.34abc

Nocellara Messinese ($37.77 \pm 23.36 \text{ mg}\cdot\text{kg}^{-1}$), about 3.45 times less than in Ottobratica. The total FALS found in Picholine cultivated in South West Calabria ($48.80 \pm 16.39 \text{ mg}\cdot\text{kg}^{-1}$) were around the lower limit of the total FAL content revealed in Picholine Marocaine and Picholine Languedoc found in olive oil produced in Morocco with an FAL content ranging between $46.0 \text{ mg}\cdot\text{kg}^{-1}$ and $96.0 \text{ mg}\cdot\text{kg}^{-1}$ (El Antari *et al.*, 2000).

Similarly for Calabrian olive oils, Krichène *et al.* (2010) have found C_{26-OH} to be the most common FAL in virgin olive oil from Jdallou, Chemlali Sfax, Swabâa, El Hor, and Oueslati monovarietal Tunisian cultivars. Also Strabbioli *et al.* (2007) have found C_{26-OH} to be the greatest FAL component in the olive oil of cultivars grown in Central Italy, including the Leccino cultivar. Moreover, hexacosanol was found to be prominent in Coratina from the Apulia Region in the Southeast of Italy, in Koroneki from Crete (Aparicio and Luna, 2002), in Manzanilla cv., Picual cv. and Gordal cv. in the region of the Guadalquivir valley, Encinarejo (Córdoba - Spain), (Orozco-Solano *et al.*, 2010) and in pomace olive oil (Fernández-Arche *et al.*, 2009).

In the olive oil from cultivars grown in Central Italy, Ranalli *et al.* (2002) found much higher results ($500 \text{ mg}\cdot\text{kg}^{-1}$ in total FALS) than were found for the olive oils produced in Southwest Calabria. In the olive oil of Coratina grown in Apulia, Aparicio and Luna (2002) found a total FAL content of $63 \text{ mg}\cdot\text{kg}^{-1}$ (after two phase extraction) and $58 \text{ mg}\cdot\text{kg}^{-1}$ (after three phase extraction): less than for the Coratina oil of Southwest Calabria (83.33 ± 39.85).

3.2. Analysis of variance

The one-way ANOVA results are reported in Table 1. Significant differences ($p \leq 0.05$) among cultivars were found for several fatty alcohols. Ottobratica differs significantly for C_{22-OH}. With regards to C_{24-OH} there were significant differences among Ottobratica, Pendolino and Coratina, Itrana, Nocellara Messinese, Nociara, Picholine. In the case of C_{28-OH} there were significant differences among Coratina, Ottobratica, Sinopolese and Cassanese, Itrana, Leccino, Nocellara Messinese, Nociara, Picholine. With regards to total FAL contents there were significant differences among Ottobratica, Pendolino, Sinopolese and Itrana, Nocellara Messinese, Nociara, Picholine. Cassanese had the highest significant OCFAL content. Tricosanol, pentacosanol, hexacosanol, heptacosanol, octacosanol, total FALS and ECFALS/OCFALS did not differ significantly for Ottobratica and Sinopolese, two autochthonous cultivars, meaning these two cultivars were the most similar with regards to their fatty alcohol content.

The results of two-way ANOVA are reported in Table 3. Cultivars showed a highly

TABLE 2. Percentage compositions in fatty alcohols of the different cultivars. The values represent the mean of fifteen samples

	Cassanese	Coratina	Itrana	Leccino	Nocellara					
					Messinese	Nociara	Ottobratica	Pendolino	Picholine	Sinopolese
C _{22-OH}	10.88	6.32	15.05	13.11	16.04	12.08	15.51	12.00	12.83	11.99
C _{23-OH}	2.31	1.29	1.92	1.66	3.01	2.38	2.48	0.93	1.96	2.91
C _{24-OH}	26.62	14.84	24.23	25.14	20.79	21.65	25.25	31.13	24.36	21.78
C _{25-OH}	10.13	4.46	4.24	3.27	4.63	4.29	3.33	3.86	3.76	3.84
C _{26-OH}	33.19	46.08	30.38	36.80	30.36	30.13	27.34	33.89	33.02	37.06
C _{27-OH}	3.63	2.29	2.09	2.06	2.30	3.93	2.58	1.60	2.03	2.38
C _{28-OH}	13.23	24.72	22.09	17.97	22.88	25.54	23.52	16.58	22.05	20.02
Σ ECFALs	83.92	91.96	91.74	93.01	90.07	89.39	91.61	93.61	92.25	90.82
Σ OCFALs	16.07	8.04	8.26	6.99	9.93	10.61	8.39	6.39	7.75	9.13
ECFALs/OCFALs	5.22	11.44	11.11	13.30	9.07	8.42	10.92	14.65	11.90	9.75

TABLE 3. Fatty alcohols with significant differences. ANOVA experiment: cultivar, harvest year, cultivar × harvest year. *** ($p \leq 0.001$); ** ($p \leq 0.01$); * ($p \leq 0.05$); n.s., not significant ($p > 0.05$). Each result is calculated as the mean of five different replicates for each harvest year

Fatty alcohols	Cultivar	Harvest year	Cultivar X Harvest year
C _{22-OH}	***	**	**
C _{23-OH}	**	n.s.	***
C _{24-OH}	***	n.s.	***
C _{25-OH}	***	n.s.	***
C _{26-OH}	***	n.s.	n.s.
C _{27-OH}	**	n.s.	n.s.
C _{28-OH}	***	n.s.	n.s.
Total Fatty Alcohols	***	n.s.	n.s.
ECFAs	***	n.s.	n.s.
OCFAs	***	n.s.	***
ECFAs/OCFAs	**	**	**

significant effect ($p \leq 0.01$) for tricosanol, heptacosanol and for ECFALs/OCFALs and a very highly significant effect ($p \leq 0.001$) for all other parameters. Harvest year had a highly significant effect ($p \leq 0.01$) for docosanol and for ECFLAs/OCFALs and no significant effect ($p > 0.05$) for all other FALs singularly considered, ECFALs, OCFALs and for ECFALs/OCFALs. Cultivar x harvest year had a highly significant effect ($p \leq 0.01$) for docosanol and ECFLAs/OCFALs, and a very highly significant effect ($p \leq 0.001$) for tricosanol, tetracosanol, pentacosanol and OCFALs, and no significant effect ($p > 0.05$) for hexacosanol, heptacosanol, octacosanol, total FALs or ECFALs.

3.3. Bar graphs

The C_{22-OH} contents in the three studied harvest years are reported in Figure 1. Leccino showed the maximum content in 2005 and a lowest content in the two subsequent harvest years, approximately 50% less. Pendolino had an increasing trend from 2005 to 2007. All other cultivars showed the highest contents in 2006.

The concentration in C_{23-OH} is depicted in Figure 2. Cassanese, Itrana and Nociara had an increasing trend from the first to the third harvest year. No tricosanol content or trace amounts were found in Pendolino or in Sinopolese in the second harvest year and in Picholine in the first harvest year. Coratina, Leccino and Ottobratica had the lowest concentration in 2006.

The concentration in C_{24-OH} is depicted in Figure 3. Leccino confirmed the trend found for C_{22-OH}; Cassanese and Pendolino had an increasing trend from 2005 to 2007, whereas tetracosanol in Nociara and Ottobratica decreased over the three harvest years. In the remaining cultivars the maximum content was in 2006.

The concentration in C_{25-OH} is depicted in Figure 4. The pentacosanol content increases from 2005 to 2007 in Cassanese, Itrana, Pendolino and Picholine, whereas in Nociara and Sinopolese it decreases. In Coratina and Leccino the highest content was in 2006.

The C_{26-OH} content is shown in Figure 5. Hexacosanol increased from 2005 to 2007 in Cassanese, Nociara, Ottobratica, Pendolino and Picholine. Only Coratina had a decreasing trend.

The variation in C_{27-OH} content is compared in Figure 6. The maximum accumulation for Leccino, Nociara, Ottobratica and Pendolino was found in 2006. Heptacosanol decreased in Coratina over the three studied harvest years.

The C_{28-OH} content is presented in Figure 7. Itrana, Ottobratica, Pendolino and Picholine had

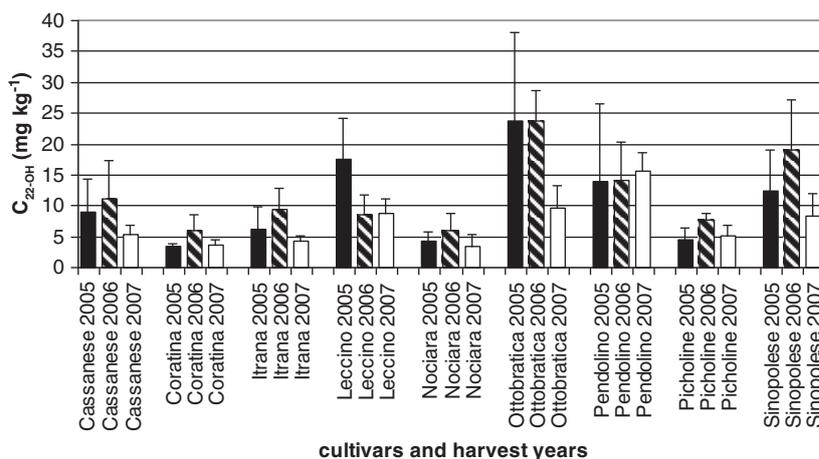


FIGURE 1. Variation in C_{22-OH} content for three harvest years 2005, 2006 and 2007, for the different cultivars. The values represent the means of five measurements ± standard deviation.

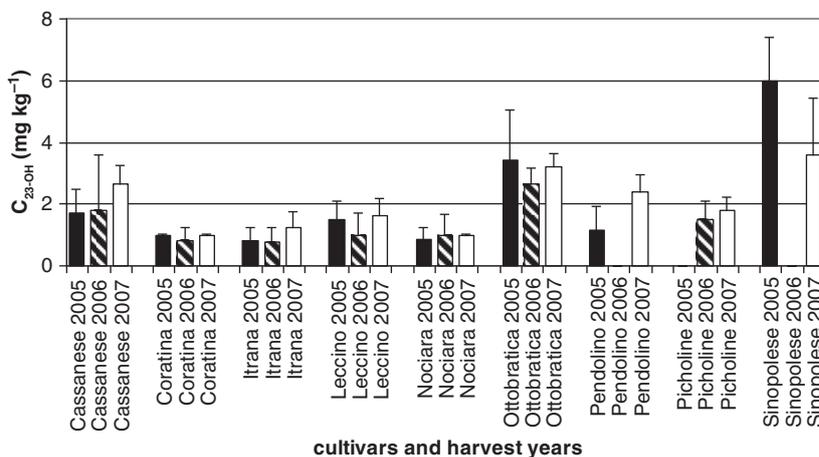


FIGURE 2. Variation in C_{23-OH} contents for three harvest years 2005, 2006 and 2007, for the different cultivars. The values represent the means of five measurements ± standard deviation.

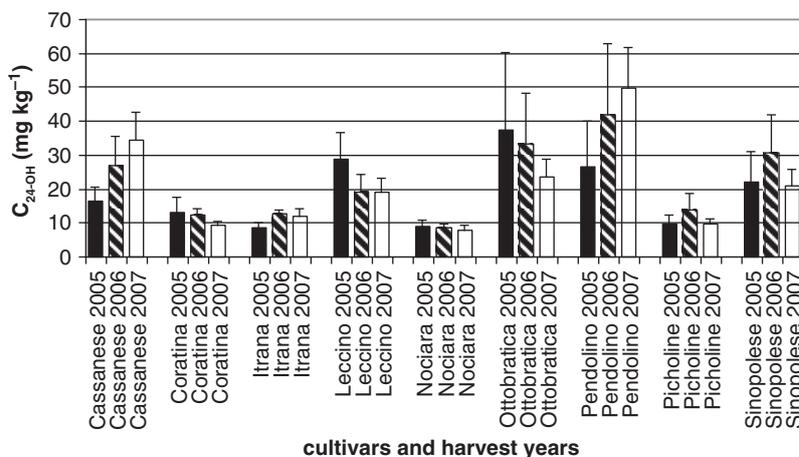


FIGURE 3. Variation in C_{24-OH} contents for three harvest years 2005, 2006 and 2007, for the different cultivars. The values represent the means of five measurements ± standard deviation.

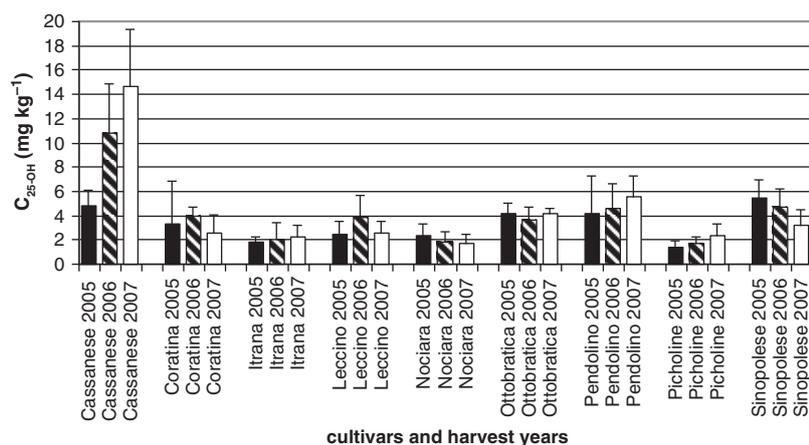


FIGURE 4. Variation in C_{25-OH} contents for three harvest years 2005, 2006 and 2007, for the different cultivars. The values represent the means of five measurements ± standard deviation.

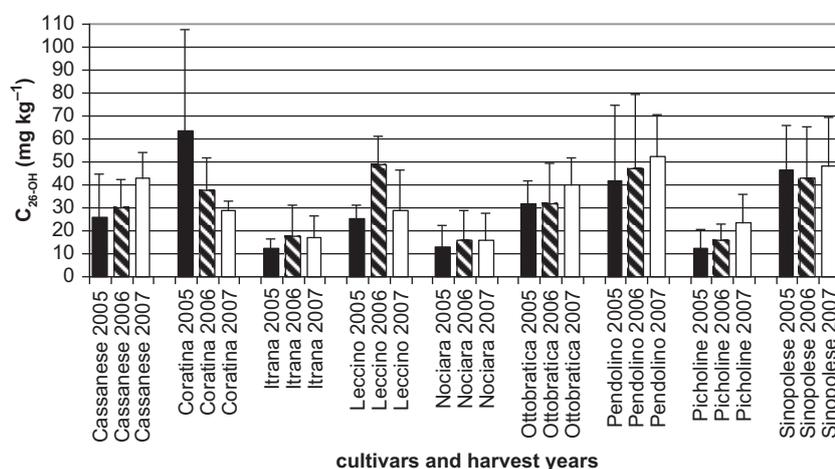


FIGURE 5. Variation in C_{26-OH} contents for three harvest years 2005, 2006 and 2007, for the different cultivars. The values represent the means of five measurements ± standard deviation.

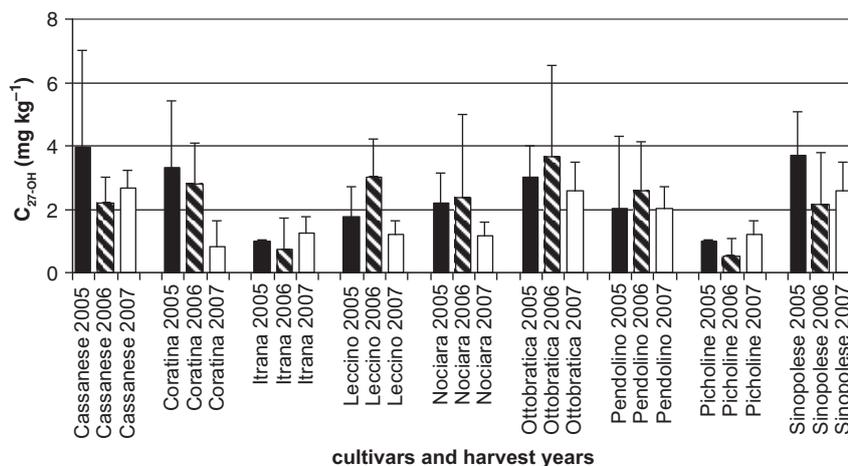


FIGURE 6. Variation in C_{27-OH} contents for three harvest years 2005, 2006 and 2007, for the different cultivars. The values represent the means of five measurements ± standard deviation.

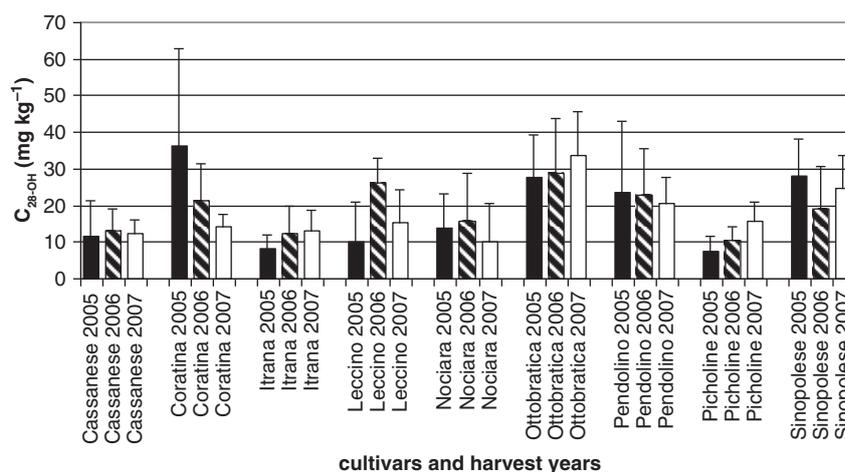


FIGURE 7. Variation in $C_{28:OH}$ contents for three harvest years 2005, 2006 and 2007, for the different cultivars. The values represent the means of five measurements \pm standard deviation.

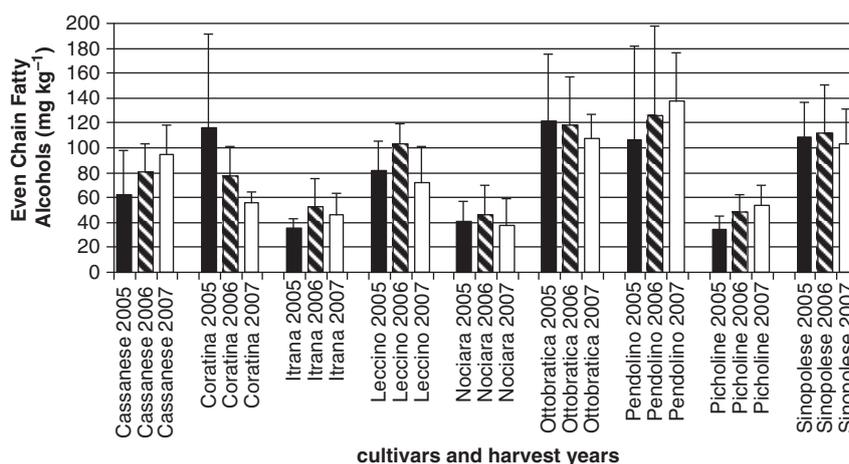


FIGURE 8. Variation in the sum of Even Chain Fatty Alcohol contents for three harvest years 2005, 2006 and 2007, for the different cultivars. The values represent the means of five measurements \pm standard deviation.

the same increasing trend found for hexacosanol and Coratina had the same decreasing trend. In Cassanese, Leccino and Nociara the highest octacosanol content was detected in 2006.

The variations in ECFALs and OCFALs are reported in Figures 8 and 9. The ECFAL and OCFAL contents increased in Cassanese, Pendolino and Picholine whereas it decreased in Coratina and Ottobratica during the three studied years. Consequently it can be seen that a similar trend was found for total FAL content (Figure 10).

In almost all cultivars the major value regarding the ECFALs/OCFALs ratio was found in 2006 (Figure 11).

CONCLUSIONS

The total amount of FALs determined in olive oils extracted from drupes of cultivars grown in Southwest Calabria (Southern Italy) was always well under $300 \text{ mg}\cdot\text{kg}^{-1}$, allowing these oils to be classified as extra virgin, according to both EU and I.O.C. regulations. The FAL data for the allochthonous cultivars show a good adaptation to the microclimatic conditions of Southwest Calabria. FAL content in olive oil was highly influenced by cultivars. The combination of cultivar and harvest year showed an effect only in some cases. By and large, the harvest year had no effect on the FAL composition.

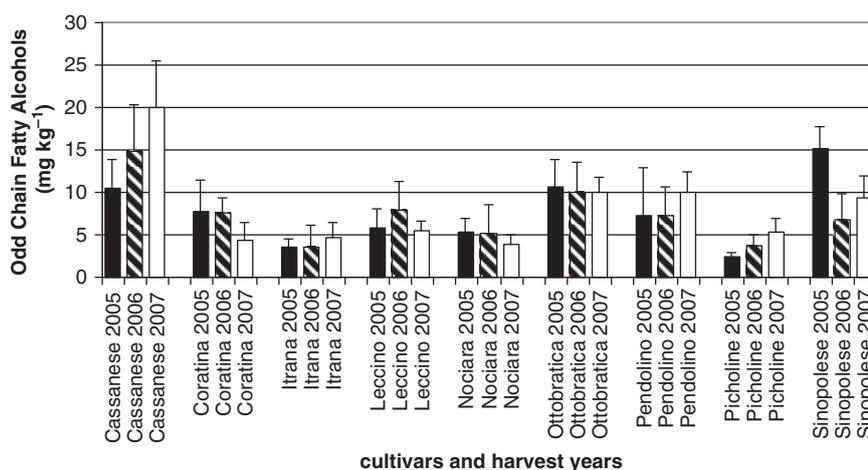


FIGURE 9. Variation in the sum of Odd Chain Fatty Alcohol contents for three harvest years 2005, 2006 and 2007, for the different cultivars. The values represent the means of five measurements±standard deviation.

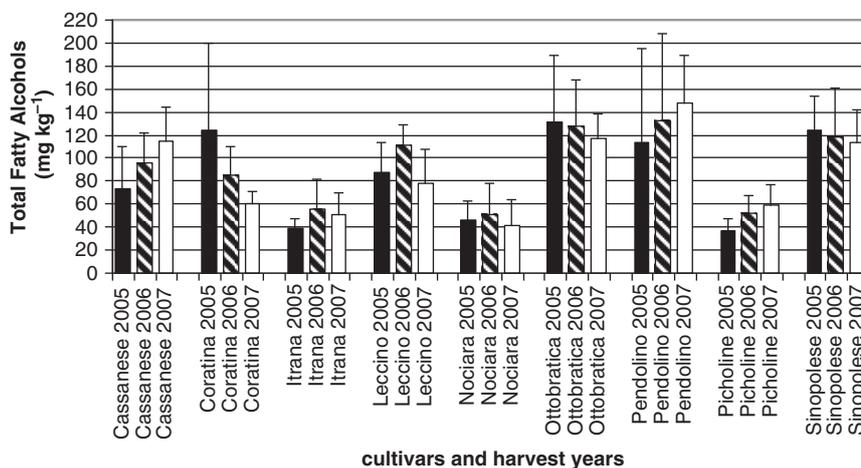


FIGURE 10. Variation in Total Fatty Alcohol contents for three harvest years 2005, 2006 and 2007, for the different cultivars. The values represent the means of five measurements±standard deviation.

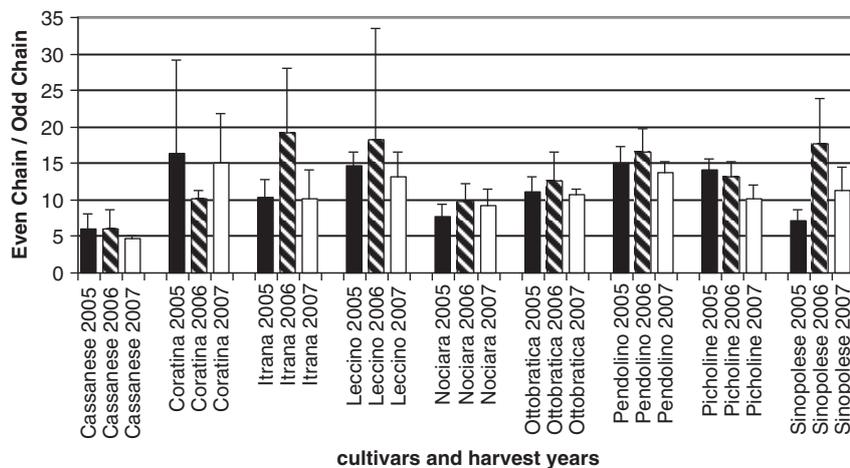


FIGURE 11. Variation in Even Chain Fatty Alcohol contents and Odd Chain Fatty Alcohol content ratio for three harvest years 2005, 2006 and 2007, for the different cultivars. The values represent the means of five measurements±standard deviation.

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