Influence of Fertigation in ‘Manzanilla de Sevilla’ Olive Oil Quality

Ana Morales-Sillero¹ and R. Jiménez

Departamento de Ciencias Agroforestales, Universidad de Sevilla, 41013 Sevilla, Spain

J.E. Fernández and A. Troncoso

Instituto de Recursos Naturales y Agrobiología (IRNAS-CSIC), Apartado 1052, 41080 Sevilla, Spain

G. Beltrán

Estación de Olivicultura y Elaiotecnía CIFA ‘Venta del Llano’, Instituto Andaluz de Investigación y Formación Agraria, Pesquera y de la Producción Ecológica (IFAPA), CICE, Junta de Andalucía, Apartado 50, 23620 Mengíbar, Jaén, Spain

Additional index words. *Olea europaea*, virgin olive oil, nutrition, nitrogen, potassium, polyphenol, oxidative stability, fatty acids

Abstract. We report the results of a study carried out in a ‘Manzanilla de Sevilla’ olive orchard near Sevilla, Spain, where the influence of different fertigation treatments on oil chemical composition was considered. Four treatments were established: control (no fertilizer) and T200, T400, and T600 in which each tree, respectively, received 200, 400, or 600 g N per irrigation season of a 4N–1P–3K complex fertilizer applied daily from 1999 to 2003. Results shown here correspond to the last 2 years of the experiment, 2002 and 2003. Fruits were sampled at the beginning of ripeness at the “green” stage. Fruit water content increased with the amount of fertilizer, probably because of the increase of potassium in the pulp. Oil content was unaffected by the treatments, but oil yield increased with the fertilizer dose in 2003 as a result of the number of fruits per tree. Polyphenol content, which is related to antioxidant oil capacity, K₂₂₅ (bitterness), and oxidative stability were lower in the oils made from trees receiving greater fertilizer doses. The monounsaturated fatty acid content, in particular oleic acid, decreased with increasing amounts of applied fertilizers, whereas polyunsaturated fatty acids, in particular linoleic acid, increased with it.

Olive oil is a basic constituent of the Mediterranean diet. Consumption has significantly increased in the last years as a result of its nutritional value and recognized benefits for human health (Martínez-Victoria and Mañas, 2004; Wahlqvist and Kouris-Lazos, 1996).

Factors such as cultivar, weather and soil conditions, fruit ripeness, agronomic practices, and oil extraction process modify oil chemical composition and organoleptic characteristics (García et al., 1996; Salvador et al., 2001; Uceda et al., 2004). Within the agronomic practices, particular attention is being paid to irrigation and, more recently, fertilization.

Additional water supplies have positive effects on fruit yield (d’Andria et al., 2004; Michelakis et al., 1995; Pastor et al., 2005). Oil content does not always increase with irrigation (Marsilio et al., 2006; Patumi et al., 2002), although oil quality is usually modified. In particular, polyphenol content, K₂₂₅ (bitterness), and oxidative stability have been observed to decrease with the increase in applied water (Berenguer et al., 2006; Magliulo et al., 2003; Patumi et al., 1999; Salas et al., 1997). The olive oil antioxidant activity is principally the result of the high polyphenol content, particularly orthodiphenols (Baldioli et al., 1996). Phenolic compounds are part of the polar fraction of virgin olive oils (García et al., 2003) and they are related to the pungent astringency and bitter taste (Gutiérrez et al., 1977; Tsimidou et al., 1992). Like other food products, these compounds have interest as a result of their potential for human health (García et al., 2003; Parr and Bolwell, 2000). A high correlation between polyphenol content and K₂₂₅ extinction coefficient has been shown (Beltrán et al., 2000). The oxidative stability parameter allows us to estimate the oil susceptibility to oxidative degeneration and is positively correlated with polyphenol con-

tent, tocopherol content, fatty acid composition, carotenoid, and chlorophyll pigments (Aparicio et al., 1999; Baldioli et al., 1996; Beltrán et al., 2000; Tsimidou et al., 1992; Vázquez et al., 1973). The polyphenol content–stability correlation is greater in oils from green than from black fruits (Caponio et al., 2001).

Many authors observed that irrigation did not affect fatty acid composition of olive oil (Berenguer et al., 2006; d’Andria et al., 2004; Motilva et al., 2000; Patumi et al., 2002). However, lower values of the monounsaturated/polyunsaturated ratio (Pastor et al., 2005) and of the unsaturated/saturated ratio (Stefanoudaki et al., 2001) were found in oil from irrigated olive trees than that from nonirrigated trees.

The widely accepted assumption that fertilization practices have little if any effect on oil quality (Fiorino et al., 1996; Uceda et al., 2004) is not supported by results from recent studies. Thus, Fernández-Escobar et al. (2006) found that nitrogen overfertilization in ‘Picual’ olive trees induced a decrease in polyphenol content and, in consequence, both in K₂₂₅ and oxidative stability of virgin olive oil. A significant decrease in saturated fatty acids content, and so an increase in unsaturated/saturated and polyunsaturated/saturated acid ratios, was observed when high levels of N and K were supplied to the soil around the tree trunks in a ‘Carrasqueña’ olive orchard (Simões et al., 2002). On the contrary, no differences in oil content and quality were found when N and K foliar applications were made on ‘Carolea’ olive trees (Inglese et al., 2002).

It has been observed that an excess of fertilizer applied by fertigation decreased fruit quality in other fruit tree species. In particular, high N doses caused thicker peels in orange (Dasberg et al., 1983) as well as later development of color in orange, grapefruit, and apple (Dasberg et al., 1983; He et al., 2003; Klein et al., 1989). In olive, despite the general practice of this method in intensive orchards, little is known about the effect on oil quality. No significant differences in oil chemical composition were found in young ‘Empeltre’ olive trees after 3 years of different nitrogen treatments (Alcubilla et al., 2002).

The aim of this work was to study the influence of N–P–K fertigation on ‘Manzanilla de Sevilla’ olive oil yield and composition. This cultivar is particularly appreciated for table olive production because of its productivity and fruit quality. Although the fruit oil content is not very high, it is appreciated because of its quality and stability (Barranco et al., 2005). Consequently, most of the production goes to table olive processing, but a substantial part may go to olive oil extraction industries when the market value for oil is high.

Materials and Methods

Plant material. The trial was carried out between 1999 and 2003 in a ‘Manzanilla de

Received for publication 22 Nov. 2006. Accepted for publication 25 Feb. 2007.

This work was supported by the *Programa de Mejora de la Calidad de la Producción del Aceite de Oliva* (Olive Oil Quality Improvement) CAO 98-004.

We thank Ignacio Girón, Ricardo Fernández-Escobar, José Ordovás, and Javier Troncoso. We also thank the staff of the Estación de Olivicultura y Elaiotecnía CIFA ‘Venta del Llano,’ Junta de Andalucía, in particular, Antonia Fernández, M^a Paz Aguilera, Maite Ruano, and Marino Uceda.

¹To whom reprint requests should be addressed; e-mail amorales@us.es

Sevilla' olive orchard located in Alcalá de Guadaira, close to Seville, Spain. The trees, planted at 7 × 7 m, were 10 years old in 1999. The soil is a Calcic Rhodoxeralf, sandy clay loam at the first 0.35 m and sandy loam or loam below this depth. The pH was ≈8.2; CaCO₃ increased from ≈30% at the top 0.35 m to 70% below 0.55 m, whereas organic matter decreased from 2.5% to 0.29%. The P and K contents were low (4 and 94 mg·kg⁻¹, respectively) at the start of the experiment. Total N content was 0.16%.

A randomized complete block design with six blocks and four trees per elementary plot was established. Each plot was surrounded by guard trees to avoid interferences among treatments. Four fertigation levels were applied; irrigation amounts were the same for all trees, as described subsequently, but the amount of fertilizer applied was different depending on the treatment. No fertilizers were supplied to trees of the control treatment; each tree of treatments T200, T400, and T600 received 100, 200, and 400 g N per year, respectively, of a 4N-1P-3K complex fertilizer in 1999, 2000, and 2001; in 2002 and 2003, these amounts were respectively increased to 200, 400, and 600 g N per tree to account for the increase in tree size. Each tree received an additional dose of ≈60 g N per irrigation season as a result of the nitrate content in the irrigation water.

Irrigation amounts were enough to replace the crop evapotranspiration (ET_c, mm) estimated with the crop coefficient approach (Allen et al., 1998). The K_r and K_c coefficient values were those estimated for the orchard by Fernández et al. (2006) (K_r = 0.7; K_c values were 0.76 in May, 0.70 in June, 0.63 in July and August, 0.72 in September, and 0.77 in October). The calculated ET_c values for the five experimental irrigation seasons (1999 to 2003) were 233, 217, 253, 248, and 256 mm, respectively.

In all treatments, trees were irrigated daily from April–May to September–October, depending on the year, with one line per tree row containing four compensating 8 L·h⁻¹ drippers per tree 1 m apart. In the fertigation treatments, a liquid N–P–K fertilizer was injected into the irrigation system, toward the end of each irrigation event, allowing for 15 min rinse of the laterals. Equal daily doses were applied along the irrigation period. N was derived from urea, NH₄, and NO₃ (2:1:1 mixture); P from H₃PO₄; and K from KCl.

Olive trees were picked by hand in September, when the maturity index was 1 (green color). This was determined according to the pigmentation extent of the epicarp and mesocarp of the olive drupes (Beltrán et al., 2004). In 2002 and 2003, a representative sample of 3.5 kg per plot was taken to the laboratory for oil extraction and chemical analyses. All trees were sampled.

Fruit water and oil content. Fruit water content was determined by desiccation at 105 °C for 24 h of a milled fruit sample. Oil content was analyzed in the same sample by nuclear magnetic resonance (The Minispec

MQ 10; Bruker). Results were expressed as percentage of fresh and dry weight.

Fruit N, P, and K content. Fruit N, P, and K content was determined in samples that were previously dried at 80 °C for 48 h and then milled. Nitrogen content was measured by spectrophotometry in a AA-3 autoanalyzer (Bran+Luebbe, Germany) after digestion with concentrated sulfuric acid using the Kjeldahl method. P and K contents were determined by ICP-OES (ThermoJarrell mod. IRIS ADVANTEGE, MA, USA) after dry ashing (550 °C) and creating a solution of the ashes in concentrated hydrochloric acid (Walinga et al., 1995). Results were expressed as g·kg⁻¹ on a dry weight basis.

Oil extraction. For oil extraction, we used an Abencor laboratory oil mill (Abengoa, Seville, Spain) (Martínez et al., 1975). Malaxation conditions of the olive paste were 30 min at 28 °C. After decantation, oil samples were filtered and stored at –24 °C until analysis.

Oil analysis. Free acidity, peroxide value, ultraviolet absorption characteristics, and fatty acid composition were determined in oil samples following analytical methods described in Regulation EEC/2568/91 (European Union Commission, 1991). Free acidity, expressed as percentage of oleic acid, was determined with a potassium hydroxide titration. Peroxide value, expressed as meq O₂ kg⁻¹ of oil, was analyzed by iodometry. K₂₃₂ and K₂₇₀ extinction coefficients were determined from 1% solution of oil in cyclohexane measuring the absorbance at 232 nm and 270 nm in a spectrophotometer HP 8452A ultraviolet-vis (Hewlett Packard, Germany). Fatty acid methyl ester composition was determined by gas chromatography in a Perkin-Elmer Autosystem (CT, USA). Results were expressed as peak area relative percentage.

Polyphenol content was determined by extraction of an oil solution in hexane with a water–methanol mixture (60:40). The Folin-Ciocalteu reagent was added and colorimetric measurements at 725 nm were made (Vázquez et al., 1973). Results were ex-

pressed as mg·kg⁻¹ of caffeic acid. Bitterness index (K₂₂₅) was determined by extraction of the bitter compounds using C18 SPE Cartridges (Baker, J.T., Holland) and measurement of the absorbance of the extract at 225 nm with a spectrophotometer (Gutiérrez et al., 1992). Tocopherol content was analyzed using the IUPAC method 2432 (IUPAC, 1992). An oil solution with hexane:2-propanol (99.5:0.5) was analyzed by high-performance liquid chromatography using a Perkin-Elmer chromatograph. Results were expressed as mg·kg⁻¹. Resistance of oil to oxidation was measured using an automated Rancimat device 679 apparatus (Methrom Co., Basel, Switzerland) with a 2.5-g oil sample warmed to 98 °C and an air flow of 10 L·h⁻¹. Results were expressed as induction time in hours (Gutiérrez, 1989).

Statistical analysis. Data were analyzed using analysis of variance. Polynomial contrasts and regression analysis were obtained when a significant F test was observed.

Results and Discussion

The fertigation treatments clearly affected fruit olive and oil composition. Although fruit nitrogen and phosphorus contents were not modified by fertigation, potassium content showed an increase with the amount of fertilizer (Table 1). The differences between T600 and the control were 15.7% in 2002 and 16.7% in 2003. This could explain the increase in water content of the fruit with the fertilizer dose observed on both years, as shown in Figure 1; differences between T600 and the control were 6.9% in 2002 and 6.8% in 2003. It is known that osmotic adjustment takes place when potassium accumulates in vacuole cells. Active absorption of this element induces water absorption in cells and plant tissues (Mengel and Kirkby, 1987).

No differences in fruit oil content, expressed as percent dry weight, were observed between treatments (Table 1). However, when the oil contents were expressed as percentage of fresh weight, we observed that it decreased in 2002 as fertilizer dose

Table 1. Fruit N, P, and K contents, oil content, and oil yield (n = 6) from each fertigation treatment and for the last two experimental seasons.²

Year and treatment	N (g·kg ⁻¹)	P (g·kg ⁻¹)	K (g·kg ⁻¹)	Oil content		Oil yield (kg·ha ⁻¹)
				(% f.w.)	(% d.w.)	
2002						
Control	6.10	1.15	9.21	16.67	47.14	304.0
T200	7.32	0.87	9.74	14.90	46.10	399.8
T400	7.15	0.87	10.52	14.30	44.25	399.8
T600	6.92	0.88	10.92	13.90	45.29	291.7
Significance	NS	NS	NS	L***	NS	NS
CV (%)	15.2	30.7	10.8	6.9	5.8	35.3
2003						
Control	6.94	1.41	13.54	13.26	32.13	112.2
T200	7.43	1.33	13.56	14.83	35.47	183.6
T400	8.41	1.19	15.53	13.45	35.72	222.4
T600	7.67	1.28	16.24	12.55	34.20	230.5
Significance	NS	NS	L**	NS	NS	L*
CV (%)	9.2	19.5	10.2	10.4	7.8	38.7

²See text for details on the treatments.

NS, **, *** Nonsignificant or significant at P ≤ 0.05, 0.01, and 0.001, respectively.

CV = coefficient of variation; d.w. = dry weight; f.w. = fresh weight; L = linear.

Fig. 1. Fruit water content ($n = 6$) from each fertigation treatment and for the last two experimental seasons.

increased. This trend could be the result of the observed moisture increase. The higher water content in fruits from the fertigation treatments, especially T600, was shown to negatively affect oil extraction. It is known that fruits from irrigated trees require the greatest co-adjuvant additions (tale) with respect to nonirrigated trees for better processing (Pastor et al., 2005).

Oil yield increased with the amount of fertilizer in 2003 (Table 1) as a consequence of the number of fruits per tree, which showed a linear and significant increase from 970 in the control to 1719 in T600.

Polyphenol content was significantly lower in 2002 than in 2003 for all treatments (Table 2), likely because of the fruit water content, which was higher in 2002 (Fig. 1). This could be a consequence, at least in part, of the 105 mm rainfall recorded before harvesting. A decrease of polyphenol content related to a rainfall event before harvesting was also observed by Motilva et al. (2000). Polyphenols are soluble both in water and oil; therefore, significant losses of these compounds with the wastewater can occur during the oil extraction process (Rodis et al., 2002).

In 2002, polyphenol content was lower in the fertigation treatments than in the control. In 2003, a trend was observed in lower oil polyphenol content with the higher fertigation treatments of T400 and T600 compared with the control and T200. Differences

between T600 and the control were 16.0% lower in 2002 and 26.4% lower in 2003. It is known that high polyphenol content could occur in stressed olive trees, probably because of the activation of enzymes such as phenylalanine ammonia lyase, which catalyzes the synthesis of most of the phenolic compounds (Parr and Bolwell, 2000). The lack of water, for instance, has a marked effect on the accumulation of polyphenols in olive oil (Berenguer et al., 2006; Patumi et al., 1999; Salas et al., 1997; Tovar et al., 2002). In our case, all trees were irrigated to replace the crop water demand, so we assume water stress of our experimental trees was low. The observed differences between treatments on polyphenol content must be attributed to differences in mineral nutrition. In fact, reduced nitrogen applications are typically associated with an increase of phenolic compounds (Waterman and Mole, 1994, cited by Parr and Bolwell, 2000). In particular, the carbon-nutrient balance hypothesis (Bryant et al., 1983) suggests that under conditions of limited availability of nutrients, plant growth will be reduced more than photosynthesis. Under these conditions, non-structural carbohydrates will be accumulated and diverted into an enhanced production of carbon-based metabolites. This may have occurred in our control treatment in 2002 and 2003 and in the T200 treatment in 2003, in which leaf nitrogen levels were below the threshold limit for deficiency (data not shown). In a recent study conducted in two adult 'Picual' olive orchards, a decrease in oil polyphenol content was found with increased nitrogen dose applied either to soil or soil and foliage, depending on the orchard. In this case, leaf nitrogen levels were always above the threshold limit for deficiency and it showed a linear increase with the amount of fertilizer applied (Fernández-Escobar et al., 2006). The decrease in oil polyphenol content between our control and T600 treatments could be also explained, in part, by the increase in fruit water content, which could affect the extraction of partially soluble phenolic compounds as observed in a previous study by Salvador et al. (2001).

The mean values of the K_{225} extinction coefficient were high (Table 2), likely because the fruits were harvested at an immature "green" stage. The responses of both K_{225} and oil oxidative stability to the fertigation treatments were similar to that of the polyphenol content (Table 2). Several reports show the polyphenol content is responsible for the oil bitterness (Beltrán et al., 2000; Gutiérrez, 1989; Salas et al., 1997). Furthermore, the polyphenol content as well as the monounsaturated/polyunsaturated fatty acid ratio (Table 3) probably influenced the oil oxidative stability. High positive correlations between oil oxidative stability and polyphenol content have been observed by other authors (Uceda et al., 2004; Vázquez et al., 1973). Aparicio et al. (1999) also found a positive correlation between oil oxidative stability and the oleic/linoleic fatty acid ratio.

Our data show that fatty acid composition was also modified by the fertigation treatments (Tables 3 and 4). Monounsaturated fatty acids decreased as the fertilizer dose increased. On the contrary, the content of polyunsaturated fatty acids increased with it. In particular, the oleic acid content decreased with the amount of fertilizer at the same time that linoleic fatty acid content increased. The linolenic acid content was also modified, showing in general greater values for the two highest fertigation treatments. The monounsaturated/polyunsaturated fatty acid ratio decreased for the T600 treatment, as compared with the control, by 3.4% in 2002 and 5.2% in 2003. The unsaturated/saturated acid ratio was unaffected by the treatments.

These results are interesting for olive oil because the beneficial effects on human health are attributed, in part, to the high content of monounsaturated fatty acids, particularly oleic acid. In addition, a greater oleic acid content but a lower linoleic acid content seems to improve oil oxidative stability (Aparicio et al., 1999). In our study, differences between treatments in oleic and linoleic acid contents, in particular, were significant and consistent and occurred after 5 years of fertigation treatments. It is not easy, however, to find an explanation. Simões et al. (2002) found a significant decrease in saturated fatty acids content and so an increase in unsaturated/saturated and polyunsaturated/saturated fatty acid ratios when supplied to the soil around the tree trunks high levels of N and K in a 'Carrasqueña' olive orchard. On the contrary, no differences in oil content and quality were found when N and K foliar applications were made on 'Carolea' olive trees (Inglese et al., 2002). In others oleaginous species, it has been suggested that water and nutritional status may influence the genetically programmed activation or synthesis of oleate desaturase (Flagella et al., 2002).

Other differences in oil composition were found between treatments (Table 5). Free acidity increased with the amount of the applied fertilizer in 2002, whereas peroxide value and K_{232} and K_{270} extinction

Table 2. Polyphenol content, K_{225} , and oxidative stability of virgin olive oil extracted from fruit samples ($n = 6$) from each fertigation treatment and for the last two experimental seasons.^z

Year and treatment	Polyphenol content (mg·kg ⁻¹)	K_{225}	Oxidative stability (h)
2002			
Control	620	0.37	109
T200	452	0.29	89
T400	452	0.29	75
T600	521	0.31	80
Significance	Q**	Q*	L**, Q*
CV (%)	15.1	15.0	11.8
2003			
Control	1272	0.54	154
T200	1281	0.53	148
T400	794	0.41	97
T600	860	0.48	103
Significance	L***, C**	C**, L*	L***, C**
CV (%)	12.3	6.5	10.4

^zSee text for details on the treatments.

***Significant at $P \leq 0.05$, 0.01, and 0.001, respectively.

CV = coefficient of variation; L = linear; Q = quadratic; C = cubic.

Table 3. Fatty acid–relative composition (%) of olive oil extracted from fruit samples (n = 6) from each fertigation treatment and for the last two experimental seasons.^z

Year and treatment	Monounsaturated	Polyunsaturated	Unsaturated	Saturated	Ratio	
					Mono./Poly.	Unsat./Sat.
2002						
Control	73.66	6.85	80.51	19.41	11.02	4.15
T200	71.95	8.46	80.41	19.60	8.73	4.11
T400	70.94	9.46	80.40	19.61	7.58	4.11
T600	70.21	9.76	79.97	20.04	7.31	3.99
Significance	L**	L***	NS	NS	L***	NS
CV (%)	2.3	13.9	0.7	2.76	15.6	3.3
2003						
Control	74.51	5.07	79.58	20.43	14.82	3.89
T200	74.31	5.51	79.82	20.19	13.72	3.96
T400	72.40	7.41	79.80	20.20	9.93	3.95
T600	71.69	7.55	79.24	20.61	9.63	3.85
Significance	L***, C*	L***, C*	NS	NS	L***, C*	NS
CV (%)	1.1	10.1	0.6	1.9	11.7	2.4

^zSee text for details on the treatments.

NS, *, **, ***Nonsignificant or significant at $P \leq 0.05$, 0.01, and 0.001, respectively.

L = linear; C = cubic; Mono. = monounsaturated; Poly. = polyunsaturated; Unsat. = unsaturated; Sat. = saturated; CV = coefficient of variation.

Table 4. Fatty acid composition (%) of olive oil extracted from fruit samples (n = 6) from each fertigation treatment and for the last two experimental seasons.^z

Year and treatment	C16	C'16	C17	C'17	C18	C'18	C''18	C'''18	C20	C'20	C22
2002											
Control	16.11	1.86	0.13	0.26	2.61	71.30	6.28	0.57	0.46	0.25	0.11
T200	16.23	1.96	0.12	0.26	2.68	69.48	7.80	0.66	0.46	0.25	0.11
T400	16.23	2.00	0.12	0.26	2.68	68.42	8.77	0.69	0.47	0.26	0.11
T600	16.65	2.09	0.12	0.26	2.72	67.61	9.07	0.69	0.44	0.25	0.10
Significance	NS	NS	NS	NS	NS	L**	L***	L**, Q**	NS	NS	NS
CV (%)	2.8	7.8	5.6	5.4	5.7	2.6	14.6	6.2	7.3	6.9	13.1
2003											
Control	17.44	1.76	0.14	0.27	2.21	72.21	4.25	0.81	0.49	0.27	0.12
T200	17.03	1.68	0.15	0.27	2.41	72.08	4.78	0.73	0.50	0.27	0.11
T400	17.12	1.74	0.13	0.27	2.36	70.10	6.58	0.83	0.49	0.29	0.10
T600	17.37	1.64	0.14	0.29	2.30	70.02	6.34	0.87	0.46	0.29	0.07
Significance	NS	NS	NS	NS	Q*	L**, C*	L***, C*	C*, Q*	NS	NS	NS
CV (%)	2.6	13.2	6	4.5	3.1	1.2	11.1	5.5	6.5	7.1	4.4

^zSee text for details on the treatments. C16 = Palmitic; C'16 = Palmitoleic; C17 = Margaric; C'17 = Heptadecenoic; C18 = Stearic; C'18 = Oleic; C''18 = Linoleic; C'''18 = Linolenic; C20 = Arachidic; C'20 = Gadoleic; C22 = Behenic.

NS, *, **, ***Nonsignificant or significant at $P \leq 0.05$, 0.01, and 0.001, respectively.

CV = coefficient of variation; L = linear; Q = quadratic; C = cubic.

Table 5. Acidity, peroxide value, K_{232} and K_{270} coefficients extinction, and the content of tocopherols of olive oil extracted from fruit samples (n = 6) from each fertigation treatment and for the last two experimental seasons.^z

Year and treatment	Acidity (% oleic acid)	Peroxide value (meq O ₂ kg ⁻¹)	K ₂₃₂	K ₂₇₀	Tocopherols (mg·kg ⁻¹)			
					α	β	γ	Total
2002								
Control	0.3	9.1	1.50	0.15	147	3	12	162
T200	0.4	8.3	1.44	0.14	167	4	14	184
T400	0.4	8.9	1.42	0.14	149	4	14	169
T600	0.5	10.0	1.50	0.15	159	5	14	178
Significance	L***	NS	NS	NS	NS	NS	NS	NS
CV (%)	18.4	12	4.4	10.5	8.7	35.4	10	8
2003								
Control	0.4	15.0	1.78	0.23	278	12	13	304
T200	0.4	16.6	1.80	0.23	248	10	13	270
T400	0.4	13.1	1.63	0.18	241	11	14	265
T600	0.3	11.8	1.70	0.18	249	7	13	269
Significance	NS	L**	L*, C*	L**, C*	NS	NS	NS	NS
CV (%)	27.1	11.5	4.3	9.5	4	32.8	13.9	12.2

^zSee text for details on the treatments.

NS, *, **, ***Nonsignificant or significant at $P \leq 0.05$, 0.01, and 0.001, respectively.

L = linear; C = cubic; CV = coefficient of variation.

coefficients showed lower values for T400 and T600 in 2003. Oils from all treatments were always classified as extra virgin under International Olive Oil Council standards. Although K_{270} coefficient extinction values were greater than 0.20 in the control and

T200 treatments in 2003, this was probably the result of the high pigments content because oils were not passed through alumina.

Tocopherols contents, in particular α and β , were significantly greater in 2003 than in 2002, probably because of the stress incited

by the higher temperatures at summer. The fertigation treatments did not modify them. Because the oxidative stability showed a response to these, it seems that the effect of α -tocopherols as natural antioxidants was secondary in comparison with that of

polyphenol content in agreement with what has been reported by other authors (Aparicio et al., 1999; García et al., 1996).

In summary, this study confirms the importance of correct management of olive tree nutrition to produce oil with the highest quality for human nutrition and health. Our data show that N–P–K fertigation, a common practice in intensive olive orchards, can increase oil yield but also negatively affect oil quality. Fruit water content increased with the fertilizer amount, probably as a result of the increase of potassium content and after osmotic adjustment. This could negatively affect oil extraction. Besides, polyphenol content, K_{225} (bitterness), and oxidative stability were lower in the oils made from trees receiving greater fertilizer doses. We also observed a decrease of monounsaturated fatty acid content, in particular oleic acid, and an increase in polyunsaturated fatty acids content, in particular linoleic acid, with increasing amounts of applied fertilizers. Little differences were observed between T400 and T600 so, for our conditions, fertigating with doses similar to the T400 treatment could lead to the best equilibrium among olive oil quality, fertilization costs, and environmental impact.

Literature Cited

- Alcubilla, M., M.S. Gracia, M.A. Romero, J.L. Espada, J. Betrán, and E. Monge. 2002. Fertilización nitrogenada en olivo del cv. Empeltre. II. Composición polifenólica de hojas y frutos y calidad del aceite. *Actas del IX Simposio Ibérico sobre Nutrición Mineral de las Plantas*. 10–13 Sept. Zaragoza, p. 243–246.
- Allen, R., L.S. Pereira, D. Raes, and M. Smith. 1998. Crop evapotranspiration—Guidelines for computing crop water requirements—FAO irrigation and drainage paper 56, FAO, Rome. <http://www.fao.org/docrep/X0490E/X0490E00.htm>.
- Aparicio, R., L. Roda, M.A. Albi, and F. Gutiérrez. 1999. Effect of various compounds on virgin olive oil stability measured by rancimat. *J. Agr. Food Chem.* 47:4150–4155.
- Baldoli, M., M. Servili, G. Perretti, and G.F. Montedoro. 1996. Antioxidant activity of tocopherols and phenolic compounds of virgin olive oil. *J. Amer. Oil Chem. Soc.* 73:1589–1593.
- Barranco, D., I. Trujillo, and L. Rallo. 2005. *Elaiografía hispánica*, p. 45–231. In: *Variedades de olivo en España*. (Libro I). Rallo, L., D. Barranco, J. Caballero, C. del Río, A. Martín, J. Tous, and I. Trujillo (eds.). Junta de Andalucía, MAPA and Mundi-Prensa, Madrid.
- Beltrán, G., A. Jiménez, M.P. Aguilera and M. Uceda. 2000. Análisis mediante HPLC de la fracción fenólica del aceite de oliva virgen de la variedad Arbequina. Relación con la medida del amargor K_{225} y la estabilidad. *Grasas y aceites* 51:320–324.
- Beltrán, G., M. Uceda, M. Hermoso, and L. Frías. 2004. Maduración, p. 159–183. In: Barranco, D., R. Fernández-Escobar, and L. Rallo (eds.). *El cultivo del olivo*. Mundi-prensa, Madrid.
- Berenguer, M.J., P.M. Vossen, S.R. Grattan, J.H. Connell, and V.S. Polito. 2006. Tree irrigation levels for optimum chemical and sensory properties of olive oil. *HortScience* 41:427–432.
- Bryant, J.P., F.S. Chapin, and D.R. Klein. 1983. Carbon/nutrient balance of boreal plants in relation to vertebrate herbivory. *Oikos* 40:357–368.
- Caponio, F., T. Gomes, and A. Pasqualone. 2001. Phenolic compounds in virgin olive oils: Influence of the degree of olive ripeness on organoleptic characteristics and shelf-life. *Eur. Food Res. Technol.* 212:329–333.
- d'Andria, R., A. Lavini, G. Morelli, M. Patumi, S. Terenziani, D. Calandrelli, and F. Fragnito. 2004. Effect of water regimes on five pickling and double aptitude olive cultivars (*Olea europaea* L.). *J. Hort. Sci. Biotechnol.* 79: 18–25.
- Dasberg, S., H. Bielora, and J. Erner. 1983. Nitrogen fertigation in Shamouti oranges. *Plant Soil* 75:41–49.
- European Union Commission. 1991. Regulation EEC 2568/91 on the characteristics of olive oil and olive pomace and their analytical methods. *Off. J. Euro. Comm.* L248.
- Fernández, J.E., A. Díaz-Espejo, J.M. Infante, P. Durán, M.J. Palomo, V. Chamorro, I.F. Girón, and L. Villagarcía. 2006. Water relations and gas exchange in olive trees under regulated deficit irrigation and partial rootzone drying. *Plant Soil* 284:273–291.
- Fernández-Escobar, R., G. Beltrán, M.A. Sánchez-Zamora, J. García-Novelo, M.P. Aguilera, and M. Uceda. 2006. Olive oil quality decreases with nitrogen over-fertilization. *HortScience* 41:215–219.
- Fiorino, P., S. Alessandri, A. Cert, I. Dikmen, and M. Rahmani. 1996. Técnicas agronómicas y características del aceite de oliva, p. 195–222. In: *Enciclopedia mundial del olivo*. Consejo Oleícola Internacional. Ed. Plaza y Janés S.A., Barcelona.
- Flagella, Z., T. Rotunno, E. Tarantino, R. Di Caterina, and A. De Caro. 2002. Changes in seed yield and oil fatty acid composition of high oleic sunflower (*Helianthus annuus* L.) hybrids in relation to the sowing date and the water regime. *Eur. J. Agron.* 17:221–230.
- García, A., M. Brenes, P. García, C. Romero, and A. Garrido. 2003. Phenolic content of commercial olive oils. *Eur. Res. Food Technol.* 216:520–525.
- García, J.M., S. Sella, and M.C. Pérez-Camino. 1996. Influence of fruit ripening on olive oil quality. *J. Agr. Food Chem.* 44:3516–3520.
- Gutiérrez, F. 1989. Determinación de la estabilidad oxidativa de los aceites de oliva vírgenes: Comparación entre el método del oxígeno activo (AOM) y el método Rancimat. *Grasas y aceites* 40:1–5.
- Gutiérrez, R., C. Janer, M.L. Janer, F. Gutiérrez, and A. Vázquez. 1977. Relación entre los polifenoles y la calidad y estabilidad del aceite de oliva virgen. *Grasas y aceites* 28:101–106.
- Gutiérrez, F., S. Perdiguer, R. Gutiérrez, and J.M. Olías. 1992. Evaluation of the bitter taste in virgin olive oil. *J. Amer. Oil Chem. Soc.* 69:394–395.
- He, Z.L., D.V. Calvert, A.K. Alva, D.J. Banks, and Y.C. Li. 2003. Thresholds of leaf nitrogen for optimum fruit production and quality in grapefruit. *Soil Sci. Soc. Amer. J.* 67:583–588.
- Inglese, P., G. Gullo, and L.S. Pace. 2002. Fruit growth and olive oil quality in relation to foliar nutrition and time of application. *Acta Hort.* 586:507–509.
- IUPAC. 1992. Standard method for the analysis of oils, fats and derivatives. Method 2432. Pergamon Press, Oxford.
- Klein, I., I. Levin, B. Bar-Yosef, R. Assaf, and A. Berkovitz. 1989. Drip nitrogen fertigation of 'Starking Delicious' apple trees. *Plant Soil* 119:305–314.
- Magliulo, V., R. d'Andria, A. Lavini, G. Morelli, and M. Patumi. 2003. Yield and quality of two rainfed olive cultivars following shifting to irrigation. *J. Hort. Sci. Biotechnol.* 78:15–23.
- Marsilio, V., R. d'Andria, B. Lanza, F. Russi, E. Iannuci, A. Lavini, and G. Morelli. 2006. Effect of irrigation and lactic acid bacteria inoculants on the phenolic fraction, fermentation and sensory characteristics of olive (*Olea europaea* L. cv. Ascolana Tenera) fruits. *J. Sci. Food Agr.* 86:1005–1013.
- Martínez, J.M., E. Muñoz, J. Alba, and A. Lanzón. 1975. Informe sobre la utilización del analizador de rendimientos Abencor. *Grasas y Aceites* 26:379–385.
- Martínez-Victoria, E. and M. Mañas. 2004. El aceite de oliva en la dieta y salud humanas, p. 733–754. In: R. Fernández-Escobar and L. Rallo (eds.). *El cultivo del olivo*. Mundi-prensa, Madrid.
- Mengel, K. and E.A. Kirkby. 1987. Principles of plant nutrition. 4th ed. International Potash Institute.
- Michelakis, N., E. Vouyoukalou, and G. Clapaki. 1995. Plant growth and yield response of olive tree cv Kalamon, for different levels of soil water potential and methods of irrigation. *Adv. Hort. Sci.* 9:136–139.
- Motilva, M.J., M.J. Tovar, M.P. Romero, S. Alegre, and J. Girona. 2000. Influence of regulated deficit irrigation strategies applied to olive trees (*Arbequina* cultivar) on oil yield and oil composition during the fruit ripening period. *J. Sci. Food Agr.* 80:2037–2043.
- Parr, A.J. and G.P. Bolwell. 2000. Phenols in the plant and in man. The potential for possible nutritional enhancement of the diet by modifying the phenols content or profile. *J. Sci. Food Agr.* 80:985–1012.
- Pastor, M., V. Vega, J.C. Hidalgo, and J. Girona. 2005. Influencia del riego sobre la calidad del aceite y sobre el comportamiento de la aceituna en el proceso industrial de extracción, p. 165–184. In: Pastor, M. (ed). *Cultivo del olivo con riego localizado*. Mundi-Prensa, Consejería de Agricultura y Pesca, Junta de Andalucía, Madrid.
- Patumi, M., R. d'Andria, G. Fontanazza, G. Morelli, P. Giorio, and G. Sorrentino. 1999. Yield and oil quality of intensively trained trees of three cultivars of olive (*Olea europaea* L.) under different irrigation regimes. *J. Hort. Sci. Biotechnol.* 74:729–737.
- Patumi, M., R. d'Andria, V. Marsilio, G. Fontanazza, G. Morelli, and B. Lanza. 2002. Olive and olive oil quality after intensive monocone olive growing (*Olea europaea* L. cv. Kalamata) in different irrigation regimes. *Food Chem.* 77:27–34.
- Rodis, P.S., V.J. Karathanos, and A. Mantzavinou. 2002. Partitioning of olive oil antioxidants between oil and water phases. *J. Agr. Food Chem.* 50:596–601.
- Salas, J., M. Pastor, J. Castro, and V. Vega. 1997. Influencia del riego sobre la composición y características organolépticas del aceite de oliva. *Grasas y aceites* 48:74–82.
- Salvador, M.D., F. Aranda, and G. Fregapane. 2001. Influence of fruit ripening on 'Cornicabra' virgin olive oil quality. A study of four successive crop seasons. *Food Chem.* 73:45–53.
- Simões, P., C. Pinheiro-Alves, A.M. Cordeiro, and M.E. Marcelo. 2002. Effect of the nitrogen and potassium fertilization on fatty acids composition and oxidative stability for 'Carrasquenha' cultivar olive oil at different harvest periods—preliminary study. *Acta Hort.* 586:337–340.

- Stefanoudaki, E., K. Chartzoulakis, A. Koutsafakis, and F. Kotsifaki. 2001. Effect of drought stress on qualitative characteristics of olive oil of cv Koroneiki. *Grasas y aceites* 52: 202–206.
- Tovar, M.J., M.J. Motilva, and M.P. Romero. 2002. Changes in the phenolic composition of virgin olive oil from young trees (*Olea europaea* L. cv. Arbequina) grown under linear irrigation strategies. *J. Agr. Food Chem.* 49:5502–5508.
- Tsimidou, M., G. Papadopoulos, and D. Boskou. 1992. Phenolic compounds and stability of virgin olive oil. Part I. *Food Chem.* 45: 141–144.
- Uceda, M., M. Hermoso, and M.P. Aguilera. 2004. La calidad del aceite de oliva, p. 657–684. In: Barranco, D., R. Fernández-Escobar, and L. Rallo (eds.). *El cultivo del olivo*. Mundi-prensa, Madrid.
- Vázquez, A., C. Janer, and M.L. Janer. 1973. Determinación de los polifenoles totales del aceite de oliva. *Grasas y aceites* 24: 350–357.
- Wahlqvist, M.L. and A. Kouris-Lazos. 1996. Nutrición y valor biológico, p. 349–354. In: *Enciclopedia mundial del olivo*. Consejo Oleícola Internacional. Ed. Plaza y Janés S.A., Barcelona.
- Walinga, I., J.J. Van der Lee, V.J.G. Houba, W. Van Vark, and I. Novozamsky. 1995. *Plant analysis manual*. Kluwer Academic Publishers. Dordrecht.