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Olive Oil Quality Decreases with Nitrogen Over-fertilization

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Abstract. **Mature 'Picual' olive (*Olea europaea* L.) trees growing in two different localities of Córdoba and Jaén provinces, southern Spain, were subjected to annual applications of 0, 0.12, 0.25, 0.50, or 1.0 kg N/tree in the Cordoba's experiment, and to 0 or 1.5 kg N/tree in the Jaén's experiment. Nitrogen was applied 50% to the soil and 50% through foliar application in Córdoba, and 100% to the soil in Jaén. Three years after the initiation of treatments, when the trees showed differences among them in nitrogen content, fruit were sampled at maturity from each experimental tree during six consecutive seasons to determine the effect of nitrogen fertilization on olive oil quality. Tree nitrogen status was always above the threshold limit for deficiency even in control trees, indicating that most treatments caused nitrogen over fertilization. Nitrogen in excess was accumulated in fruit and, consequently, polyphenol content, the main natural antioxidants, significantly decreased in olive oil as nitrogen increased in fruit. The decrease in polyphenols induced a significant decrease in the oxidative stability of the oil and its bitterness. Tocopherol content, on the contrary, increased with nitrogen application, mainly by an increase in α-tocopherol, the main component in the olive oil. No effect was found on pigment content, particularly carotenoid and chlorophyllic pigments, neither on fatty acid composition.**

Olives (*Olea europaea*) are produced for extraction of olive oil for human consumption and for processing as table olives. Many factors may affect olive quality, including cultural practices such as fertilization. However, no research has been published on the effect of mineral fertilizers on olive fruit or oil quality.

Among the different essential elements, nitrogen is the most commonly applied in olive orchards, as occurs in other fruit tree species, because olives usually need N in greater amounts than other mineral nutrients (Fernández-Escobar, 2004). With the increased value of the olive crop and the relatively low cost of N fertilization, olive growers have increased their application of N fertilizers based on the perception that an increase in N fertilization may insure high yields. This practice, however, contributes to soil and groundwater pollution (Giménez et al., 2001), and could negatively affect fruit quality (Weinbaum et al., 1992) because olive fruit is the strongest sink for newly acquired N fertilizer in the plant (Fernández-Escobar et al., 2004).

Many studies conducted in fruit trees have been focused on the effect of fertilizers on characteristics of the fruit such as fruit size or fruit color, and only a few works deal with the effect on the chemical composition of the fruit. These characteristics are of great importance in storage management of fruit or their products and also affect organoleptic and nutritional properties. Moreover, the results obtained have been sometimes controversial. For instance, fruit quality has been reported to decrease at high N levels in apple (Raese and Drake, 1997), grapefruit (He et al., 2003), and mango (Nguyen et al., 2004), had no adverse effects in blackberry (Alleyne and Clark, 1997) and had a positive effect in clingstone peaches (Olienyk et al., 1997). Nothing is known in the olive.

Virgin olive oil is obtained directly from the olives by applying physical methods, and presents some nutritional and sensorial characteristics that distinguish it from other edible oils, which have to be refined for consumption. Because of its high oleic acid content, olive oil is considered as a monounsaturated fat, showing a medium linoleic acid percent (Uceda et al., 2004).

Virgin olive oil contains a group of minor compounds with a high antioxidant activity. Among these compounds can be highlighted the phenolic compounds, which have nutritional effects, protect the oils against oxidation and are related with some sensorial characteristics. A high correlation has been described between the phenol content and the oil oxidative stability (Gutiérrez et al., 1977; Gutfinger, 1981). Moreover, these compounds are responsible for the bitterness and pungency of oils (Andrewes et al., 2003).

Vitamin E is another important antioxidant that in virgin olive oils is formed by α-, β- and γ-tocopherol. Usually, vitamin E activity of virgin olive oil is higher than those described for other edible oils (Beltrán et al., 2005). Pigments are responsible for oil color that is considered as a quality parameter (Mínguez et al., 1991). In virgin olive oil can be found carotenoids and chlorophyllic pigments. Carotenoids have antioxidant activity protecting oil from autoxidation, whereas chlorophyll acts as prooxidant in light (Fakourelis et al., 1987).

Composition of virgin olive oil is affected by several agronomical factors such as cultivar, fruit ripeness, agroclimatic conditions, and some growing techniques (Uceda et al., 2004). Fertilization and pruning seem to have no effect on regulated oil quality parameters such as acidity, peroxide value and UV-vis absorption (Ferreira et al., 1978). However, we are unaware of references about the influence of N fertilization on oil composition.

The purpose of the present work was to study the effect of N fertilization on olive oil quality, particularly the effect on fatty acids composition and antioxidant compounds.

Materials and Methods

Mature, nonirrigated 'Picual' olive trees growing in two different locations in Spain—the Experimental Farm of La Mina located at Cabra, southern Córdoba province, and the Experimental Farm of Venta del Llano, located at Mengibar, Jaén province—were selected in 1994 for the experiments. Trees located at Cabra, spaced 7 × 7 m apart, were arranged in a randomized block design with four blocks and five treatments. Treatments consisted of the annual application of 0, 0.12, 0.25, 0.50, or 1.0 kg N/tree, 50% applied to the soil in March and 50% through foliar application from April to October depending upon the amount of N to be applied. Trees located at Mengibar, spaced 12 × 12 m apart, were also arranged in a randomized block design with four blocks and two treatments that consisted of the annual application to the soil of 0 or 1.5 kg N/tree. In all cases the experimental unit was composed of four trees surrounded by guard trees. Urea was used as the source of N. A concentration of 4% was used when urea was foliar applied.

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Organic matter in both localities ranged from 0.8% to 1.0%.

A sample of 2 kg of fruit per experimental tree was taken at harvest to determine oil quality. Fruit samples were collected with a maturity index ranged from 3 to 4 on a scale of 0 to 7. These indexes correspond to the end of coloring of ripening fruit (Beltrán et al., 2004). In Cabra's experiment fruit samples were taken only from treatments 0 and 1 kg N/tree in 1997, 1998 and 2000 and from all treatments in 2001 and 2002. In Mengibar's experiment fruit were collected from 1998 to 2001 from both treatments.

Nitrogen removed by the crop was estimated from fruit samples of 2 kg per plot taken at harvest. Fruit were washed and rinsed. Pulp and stone were removed separately from each fruit, dried at 80 °C for 48 h, ground and stored until analysis. Nitrogen was determined by combustion, using a FP-428 Leco analyzer.

Oil extraction has been carried out using a laboratory oil mill Abencor (Abengoa, Seville). The equipment was formed by a hammer mill, a thermobeater and a vertical paste centrifuge (Martínez et al., 1975). The kneading conditions of olive paste were 30 min at 28 °C. The oily must was decanted, filtered and stored at –24 °C until analysis.

Fatty acid methyl ester (FAME) composition was determined according the EU Regulation 2568/91 (European Union Commision, 1991). Chromatographic separation was performed using a Perkin Elmer Autosystem gas chromatograph with a split–splittless injector and a FID detector, equipped with a BPX 70 capillary column of 50 m in length, 0.22 mm i.d, and 0.25-µm of film thickness (SGE, Australia). The oven temperature was 198 °C and helium was used as carrier gas. The results were expressed as peak area relative percent

Polyphenol content was analyzed by mean liquid–liquid extraction with 60 water : 40 MeOH, using Folin-Ciocalteau reagent and colorimetric measurement at 726 nm (Vázquez-Roncero et al., 1973). Results were given as mg of caffeic acid per kilogram of oil. Bitterness index (K_{225}) was determined by extraction of bitter compounds by mean SPE cartridges C18 (J.T.Baker, Spain) and measurement of the extract absorbance at 225 nm (Gutiérrez-Rosales et al., 1992). Pigment content was determined using the spectrophotometric method described by Mínguez et al. (1991); 7.5 g of oil were weighted, dissolved in cyclohexane and taken to a final volume of 25 mL, the absorbance at 670 and 470 nm were measured for chlorophyllic and carotenoid pigments determination. For all the spectrophotometric measurements a Hewlett Packard 8452A diodearray spectrophotometer was used.

Tocopherol content in oil was determined by HPLC applying the IUPAC method 2432 (IUPAC, 1992). A HPLC Perkin Elmer was used for chromatographic separation. The chromatograph was equipped with an isocratic pump Lc200 and an UV-vis detector Lc295, using a Lichrosphere Si 60 column (Merck, Spain) of 250 mm length, 4.6 mm id. and particle size of 5 µm. A solution 0.5% of 2-propanol in n-hexane was used as mobile phase, with a flow rate of 1 $mL \cdot min^{-1}$. The detection was performed at 295 nm.

An automated Metrohm Rancimat 679 apparatus (Metrohm, Switzerland) operating at 98 °C was used to determine the oxidative stability. Oil sample (2.5 g) was weighted into the reaction vessel. The conductimetry cells were filled with deionised water up to 90 mL, and the air was flown through the heated oil at 10 $L \cdot h^{-1}$. Determinations were made by duplicate and results were expressed as induction time in hours (Gutiérrez, 1989).

Analyses of variance were performed on the data to compare the effect of the treatments. All percentage values were transformed using the arcsin of the square root before analysis. Where a significant F test was observed, polynomial contrasts were obtained when more than two treatments were compared.

Results and Discussion

Leaf N concentration, a measure for the diagnosis of tree N status, was always above the threshold limit for deficiency of 1.4 % dry weight in all places and years, and in all treatments, including the untreated control, showing a general tendency to increase with the amount of N applied (data not shown). This indicates that N over fertilization was produced in all treatments except the control. As a result of this over fertilization, N accumulated in fruit (Tables 1 and 2). Although sometimes the difference between treated and untreated trees was nonsignificant, the relationship between N removed by the crop and the annual amount of N applied showed a significant linear response when the number of treatments was increased, as occurred in the Cabra's experiment. These results are in agreement with those obtained previously, indicating that the fruit are a strong sink for N in the tree (Fernández-Escobar et al., 2004).

The increased content of N in the tree and, particularly, in the olive fruit affected some parameters involved in the olive oil quality. Polyphenol content, a component of great importance in oil quality because of its antioxidant effects, significantly decreased in both places and in most of the years of study as N application increased, and provoked often a significant decrease in oil stability and bitterness (Tables 3 and 4). These oil characteristics are directly related with polyphenol content (Gutiérrez et al., 1977; Gutfinger, 1981). The data are in agreement with Delgado et al. (2004), that found an accumulation of polyphenols in the skin of grapes when no N was added in the fertilization program. High phenol content in virgin oil has been related to stress caused by water deficit conditions (Uceda et al., 2004). Because of the higher rainfalls registered in 1998 in Jaén, oils showed in this year lower phenol concentrations, as showed in Table 4.

The decrease in polyphenol content with increasing N availability in fruit may be explained by several hypothesis that explain the relationship among fertilization, plant growth rate and plant phenol content: the carbon-nutrient balance (CNB) hypothesis (Bryant et al., 1983), the growth differentiation balance (GDB) hypothesis (Herms and Mattson, 1992), and the protein competition model (PCM) (Jones and Hartley, 1999). CNB hypothesis makes predictions about the relationships between carbon to nutrient ratio of the plant and allocation to phenolics and N-containing secondary metabolites. Under conditions of limited availability of nutrients, plant growth will be reduced more than photosynthesis and thus a surplus of nonstructural carbohydrates will accumulate. These carbohydrates can be diverted into an enhanced production of carbon-based metabolites. The GDB hypothesis suggests the inverse relationship between biomass accumulation and secondary metabolism. Since no N deficiency was obtained in the experimental trees and, consequently, no differences in yield or growth were found between olive trees treated with different N doses, as has been reported previously (Fernández-Escobar et al., 2002), these hypothesis seems not to be plausible to explain the results.

The protein competition model (PCM)

Table 1. Effect of N fertilization on N content in fruit, in the experimental farm of La Mina (Cabra).

Treatment	N removed[z] (g/kg fruit)					
(kg N/tree/year)	1997	1998	1999	2000	2001	2002
0	2.8 b	3.0	2.6 a	5.3	2.7	2.0
0.125					2.7	2.4
0.500					3.1	2.4
1.0	3.9 a	3.4	3.4 b	5.7	3.2	2.5
Significance					L**	L***
CV (%)	19	11	9	4.6	7.6	5.3

[z]Letters indicate mean separation within each year at $P \leq 0.05$, by F test. Polynomial contrasts were performed in 2001 and 2002 for the amount of N applied per tree.
,*Significant at $P \leq 0.01$ or 0.001, respectively; L = linear.

Table 2. Effect of N fertilization on N content in fruit, in the experimental farm of Venta del Llano (Mengibar).

Treatment	N removed[z] (g/kg fruit)			
(kg N/tree/year)	1998	1999	2000	2001
0	3.3	3.1	3.9 b	6.0
1.5	3.7	3.3	4.6 a	6.6
CV (%)	7	4	1.7	19.0

[z]Letters indicate mean separation within each year at $P \leq 0.05$, by F test.

postulates a more specific competition between primary and secondary metabolisms. In the case of high nutrient availability, as occurs in our experiments, phenylalanine preferentially flows into protein synthesis rather than toward synthesis of phenylpropanoids via phenylalanine ammonia lyase (PAL). In our trees fruit protein content, estimated from its N content (Norbaek et al., 2003), was higher

Table 3. Effect of N fertilization on olive oil quality, in the experimental farm of La Mina (Cabra).

Treatment	Total polyphenol[z] (ppm[y])					Bitternes (K_{225})					Stability (h)				
(kg N/tree/year)	1997	1998	2000	2001	2002	1997	1998	2000	2001	2002	1997	1998	2000	2001	2002
0	601	605 a	1138 a	577	1142	0.35 a	0.33 a	0.53 a	0.39	0.53	197 a	172 a	205 a	151 a	227
0.125				459	945			0.33	0.47					161	
0.250				458	909			0.27	0.47					153	
0.500				376	748			0.22	0.40					133	
1.0	481	362 b	793 b	280	7160.29 b	0.23 b	0.42 b	0.20	0.38	167 b	130 b	171 b	99 b	168	
Significance				L***	L***			L***	L***						L***,Q***
CV (%)	13	32	13.5	15.5	11.511	30	16.5	22.3	15.7	9	29	16.6	27.2	18.1	

[z]Letters indicate mean separation within each year at $P \leq 0.05$, by F test. Polynomial contrasts were performed in 2001 and 2002 for the amount of N applied per tree.
[y]In ppm of caffeic acid.
***Significant at $P \leq 0.001$; L = linear, Q = quadratic.

Table 4. Effect of N fertilization on olive oil quality, in the experimental farm of Venta del Llano (Mengibar).

Treatment	Total polyphenol[z] (ppm[y])				Bitterness (K_{225})				Stability (h)			
(kg N/tree/year)	1998	1999	2000	2001	1998	1999	2000	2001	1998	1999	2000	2001
0	282	1430 a	941 a	993 a	0.20	0.60 a	0.49	0.50	109	290 a	194 a	201 a
1.5	221	1136 b	788 b	724 b	0.19	0.56 b	0.44	0.43	96	232 b	161 b	153 b
CV (%)	29	17	10.6	14.3	27	10	16.0	13.2	18	13	16.4	21.9

[z]Letters indicate mean separation within each year at $P \leq 0.05$, by F test.
[y]In ppm of caffeic acid.

Table 5. Effect of N fertilization on tocopherol concentration in olive oil, in the experimental farm of La Mina (Cabra).

Treatment	Tocopherols[z] (ppm)																			
(kg N/	α					β					γ					Total				
tree/year)	1997	1998	2000	2001	2002	1997	1998	2000	2001	2002	1997	1998	2000	2001	2002	1997	1998	2000	2001	2002
0	178 b	173 b	204	150	161	3	2 b	2	6	3	15	14 b	11	9	11	196 b	188 b	217	165	175
0.125				169	161				5	3				9	11				183	175
0.250				170	153				2	2				9	10				181	165
0.500				199	159				2	2				11	10				212	171
1.0	192 a	200 a	190	227	173	3	2 a	2	6	4	16	19 a	12	11	13	211 a	221 a	204	244	190
Significance				L***	Q**				NS	Q***				NS	Q*				L***,Q*	Q**
CV (%)	10	9	5.4	7.5	5.2	18	25	13.7	20.3	13.7	16	24	12.1	13.5	16.3	9	9	5.1	6.5	5.5

[z]Letters indicate mean separation within each year at $P \leq 0.05$, by F test. Polynomial contrasts were performed in 2001 and 2002 for the amount of N applied per tree.
NS,*,**,***Nonsignificant or significant at $P \leq 0.05$, 0.01, or 0.001, respectively; L = linear, Q = quadratic.

Table 6. Effect of N fertilization on tocopherol concentration in olive oil, in the experimental farm of Venta del Llano (Mengibar).

Treatment	Tocopherols[z] (ppm)											
	α			β			γ			Total		
(kg N/tree/year)	1998	2000	2001	1998	2000	2001	1998	2000	2001	1998	2000	2001
0	232 b	207 b	203 b	2 b	2	3	14	12	8	249 b	221 b	214 b
1.5	248 a	232 a	230 a	3 a	3	3	14	14	10	265 a	248 a	243 a
CV (%)	7	3.4	6.3	30	14.4	21.5	20	11.5	8.2	7	3.4	6.1

[z]Letters indicate mean separation within each year at $P \leq 0.05$, by F test.

Table 7. Effect of N fertilization on carotenoid and chlorophyllic pigments concentration in olive oil, in the experimental farm of Venta del Llano (Mengibar).

Treatment	Carotenoid[z] (ppm)				Clhorophyllic (ppm)			
(kg N/tree/year)	1998	1999	2000	2001	1998	1999	2000	2001
0	7.8 b	6.8	7.0	9.3	7.7 b	7.3	6.4	9.7
1.5	11.5 a	6.6	8.9	11.9	14.6 a	6.5	9.4	12.9
CV (%)	34	32	16.5	15.8	53	48	30.4	20.7

[z]Letters indicate mean separation within each year at $P \leq 0.05$, by F test.

Table 8. Effect of N fertilization on carotenoid and chlorophyllic pigments concentration in olive oil, in the experimental farm of La Mina (Cabra).

Treatment	Carotenoid[z] (ppm)					Chlorophyllic (ppm)				
(kg N/tree/year)	1997	1998	2000	2001	2002	1997	1998	2000	2001	2002
0	7.0	4.0	9.4 a	8.0 b	5.7	9.3	3.7	11.4 a	6.7 b	6.7
0.125					6.4					9.5
0.250					5.3					7.9
0.500					7.5					12.8
1.0	6.6	4.4	6.9 b	15.8 a	6.3	8.3	4.1	7.9 b	27.2 a	8.7
Significance					NS					NS
CV (%)	30	44	14.2	13.1	21.6	49	69	24.4	20.2	34.6

[z]Letters indicate mean separation within each year at $P \leq 0.05$, by F test. Polynomial contrasts were performed in 2002 for the amount of N applied per tree.
[NS]Nonsignificant.

Table 9. Effect of N fertilization on fatty acids concentration in olive oil, in the experimental farm of La Mina (Cabra).

	Fatty acids (%)[z]											
Treatment (kg N/tree/year)	Palmitic C16:0	Palmitoleic C16:1	Margaric C17:0	Hepta decenoic C17:1	Stearic C18:0	Oleic C18:1	Linoleic C18:2	α-Linolenic C18:3	Arachidic C20:0	Eicosenic C20:1	Behénic C22:0	Lignocéric C24:0
1997												
0	10.2	0.87	0.029	0.067	2.08	83.0	2.55	0.51 b	0.33	0.23	0.099	0.051
1	10.3	0.90	0.035	0.087	2.04	82.7	2.61	0.54 a	0.35	0.27	0.107	0.071
CV (%)	3	6	61	43	4	1	10	8	9	16	27	62
1998												
0	10.3	1.01	0.016 b	0.087	2.76	81.3	3.53	0.46 b	0.30 b	0.21 b	0.067	---
1	10.1	0.92	0.028 a	0.068	2.67	81.7	3.32	0.51 a	0.31 a	0.23 a	0.070	---
CV (%)	5	20	59	102	10	2	20	11	4	4	11	---
2000												
0	9.7 b	0.4	0.02	0.03	1.9	84.6	2.5	0.3	0.22 a	0.13	0.06	0.03
1	10.2 a	0.4	0.02	0.03	1.9	84.3	2.4	0.3	0.21 b	0.12	0.06	0.03
CV (%)	2.9	8.3	7.1	7.5	4.3	1.1	8.0	8.6	4.4	4.3	6.2	9.2
2001												
0	10.28 b	0.56	0.04	0.06 b	2.47 a	82.88 a	2.53 b	0.46 b	0.36	0.22 b	0.11	0.05
1	10.77 a	0.52	0.04	0.07 a	2.26 b	82.07 b	2.85 a	0.60 a	0.35	0.23 a	0.11	0.05
CV (%)	2.0	14.0	6.5	7.8	3.0	0.7	3.8	5.7	2.7	2.3	4.4	9.2
2002												
0	12.24	1.05	0.03	0.07	3.19	78.1	4.0	0.45	0.35	0.21	0.09	
0.125	12.26	1.04	0.03	0.07	2.85	79.0	3.6	0.46	0.36	0.21	0.07	
0.250	12.26	1.00	0.03	0.07	2.73	79.4	3.4	0.44	0.34	0.22	0.07	
0.500	12.4	1.02	0.04	0.07	2.82	79.1	3.4	0.48	0.34	0.22	0.08	
1.0	12.44	1.08	0.03	0.08	2.89	78.5	3.9	0.47	0.36	0.21	0.08	
Significance	NS	NS	NS	L**	Q**	NS	Q***	L**	NS	NS	Q*, C*	
CV (%)	1.8	7.0	8.8	4.3	3.9	1.4	8.1	3.1	4.8	2.8	12.3	

[z]Letters indicate mean separation within each acid and year at $P \leq 0.05$, by F test. Polynomial contrasts were performed in 2002 for the amount of N applied per tree.
NS,*,**,***Nonsignificant or significant at $P \leq 0.05$, 0.01, or 0.001, respectively; L = linear, Q = quadratic, C = cubic.

Table 10. Effect of N fertilization on fatty acids concentration in olive oil, in the experimental farm of Venta del Llano (Mengíbar).

	Fatty acids (%)[z]											
Treatment (kg N/tree/year)	Palmitic C16:0	Palmitoleic C16:1	Margaric C17:0	Hepta decenoic C17:1	Stearic C18:0	Oleic C18:1	Linoleic C18:2	α-Linolenic C18:3	Arachidic C20:0	Eicosenic C20:1	Behénic C22:0	Lignocéric C24:0
1998												
0	10.1 a	1.02 a	0.02	0.07	3.1	81.8	3.2	0.52	0.36 b	0.23	0.09	0.02
1.5	9.7 b	0.94 b	0.02	0.06	3.1	81.3	3.2	0.53	0.37 a	0.24	0.10	0.03
CV (%)	3	9	75	20	14	1	15	6	4	6	11	97
1999												
0	9.0	0.56	0.0019	0.05	2.2	84.8	2.6 b	0.32	0.26	0.16	0.06 b	0.03
1.5	9.2	0.55	0.0094	0.05	2.2	85.0	2.8 a	0.33	0.26	0.16	0.07 a	0.02
CV (%)	5	16	204	9	7	2	9	10	6	5	19	79
2000												
0	9.8	0.46	---	0.03	2.4	84.4	2.1	0.29 b	0.22	0.13	0.05	0.03
1.5	9.7	0.48	---	0.03	2.5	84.2	2.4	0.31 a	0.22	0.13	0.05	0.03
CV (%)	2.7	7.9	---	5.7	6.6	1.3	8.0	2.9	2.9	2.9	11.0	9.3
2001												
0	10.7	0.79 a	0.04	0.08	2.9	81.4	2.9	0.50	0.39	0.25	0.11	0.06
1.5	10.8	0.72 b	0.04	0.09	3.0	82.9	3.0	0.51	0.38	0.24	0.11	0.05
CV (%)	2.7	5.3	9.0	11.3	4.4	0.8	3.9	5.0	4.4	3.3	7.0	11.0

[z]Letters indicate mean separation within each acid and year at $P \leq 0.05$, by F test.

whereas polyphenol content in olive oil was lower, making this hypothesis more plausible to explain these results.

Total tocopherols usually increased with N application, mainly by an increase of α-tocopherol (Tables 5 and 6), showing an inverse trend than polyphenols. Tocopherols play an important role in the quality of virgin olive oil because of its antioxidant activity, being more efficient at relatively lower concentrations. However, when α-tocopherol is present at relatively larger concentrations, a pro-oxidative effect may occur (Belizit and Grosch, 1986).

Carotenoid and chlorophyllic pigments seem to be unaltered by treatments, although sometimes these increased (Table 7, 1998) and sometimes decreased (Table 8, 2000) with N applications. Also, fatty acids were unaltered by treatments (Tables 9 and 10). Significant differences obtained between control and fertilized trees were occasional since the differences were not repeated in space or time. The only exception was α-linolenic acid that seems to increase with N applications.

In summary, N over fertilization of olive trees causes an accumulation of N in fruit that negatively affects some components of the olive oil quality. Excess of N reduces polyphenol content, the main natural antioxidants, and also the oxidative stability of the oil and its bitterness, although it seems to increase total tocopherols. No effect was found on pigments content and in fatty acid composition.

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