



Article Response of Olive Trees (*Olea europaea* L.) cv. Kalinioti to Nitrogen Fertilizer Application

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Abstract: Nitrogen is the most commonly managed mineral nutrient in olive groves because it is essential for plant growth. The precise management of N fertilization in olive cultivation is still not fully clarified, but it is essential for providing sustainable production. A nitrogen fertilizer experiment with olive trees (cv. Kalinioti) was carried out over a six-year period. Seven levels of nitrogen fertilizer given as ammonium nitrate (control, 1, 2, 3, 4, 5, and 6 kg/tree) were annually applied in order to determine the effect of nitrogen on vegetative growth, fruit set, fruit weight, yield, maturation index, and leaf N, P, and K concentrations. The results indicate that, under these conditions, application of up to 4 kg NH₄NO₃/tree significantly increased yield to 62.5 kg/tree compared to the control (37.09 kg/tree). The positive effect was attributed to the initial and final fruit set increases (7.63 and 3.73%, respectively at 4 kg NH₄NO₃/tree). However, the weight of 100 olives $(W_{100} = 331 \text{ g})$ at 4 kg NH₄NO₃/tree obtained during harvest was considerably lower compared to the control ($W_{100} = 384$ g). Higher nitrogen rates decreased yield while increasing overall shoot growth. Nitrogen fertilization did not significantly influence the oil content of olive fruit. Fruit weight, maturation index, and concentration of oil reached maximum levels in the beginning of December, indicating a suitable start to olive harvesting. The concentration of N in olive leaves increased from 1.23% to 2.38% as fertilizer levels increased from 0 to 6 kg NH₄NO₃. Maximum yield was achieved at a level of 6 kg NH_4NO_3 /tree, which corresponded to 2.01% N in leaves. The results suggest that application of 3 kg NH₄NO₃/tree can be recommended for table olive production, due to the fact that fruit weight was not decreased, while fertilization with 4 kg NH₄NO₃/tree was suitable for oil olives.

Keywords: olive fruit; yield; ammonium nitrate; leaf concentration; fruit set; oil content; maturation index

1. Introduction

Application of nitrogen is the main aspect of olive orchard (*Olea europaea* L.) fertilization due to its significant influence on plant growth as a key nutritional component [1,2]. Many studies have documented the key role of N fertilization on improving olive production [3–5]. These studies have consistently reported a gradual increase in productivity as a result of nitrogen applications.

There is some disagreement on the importance of applying nitrogen once a year to increase olive production when it comes to fertilization management, since N is often given in too large of amounts [1,6,7]. According to Fernández-Escobar et al. [8], a major problem arises in olive orchards due to excessive nitrogen fertilization, leading to various adverse effects on both the trees and the soil. Furthermore, excessive use of fertilizer faces significant costs and results in the loss of nitrogen through leaching, which has detrimental effects on the ecosystem [9]. The optimal management of nitrogen fertilization in olive orchards, including fertilizer dose and timing of distribution, remains poorly understood [6,10,11].



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Copyright: © 2023 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https:// creativecommons.org/licenses/by/ 4.0/). However, it is of paramount significance to develop management strategies aimed at mitigating the environmental consequences of nitrogen losses and minimizing the adverse impacts of excessive nitrogen on tree health.

Many works on olive nutrition have pointed out that nitrogen fertilization (a) promotes olive yield [3–5]; (b) reduces the phenomenon of alternate bearing [5]; (c) increases fruit set [12]; (d) increases shoot length and the number of inflorescences, and regulates the growth of female bodies of olive flowers [13]. Most of these studies have used between 0.5 and 1.3 kg N/tree in their experiments, while the application of excessive amounts of nitrogen had adverse effects on yield, fruit weight, fruit set, oil content, and oil quality [5,12–15]. Conversely, a lack of nitrogen in olive tree orchards leads to significant decreases in productivity [16]. Haberman [5] noted that when the concentration of nitrogen in the leaves of trees fell below 1.3%, there was a significant decrease in production. Low nitrogen availability led to decreased vegetative growth, flowering, fruit set, and ultimately fruit production. Additionally, the trees seemed to be more prone to alternate bearing. Furthermore, it has been observed that both excessive and insufficient amounts of nitrogen have a considerable negative impact on oil productivity [12].

Nitrogen fertilization is usually applied to the soil at 0.5–1.5 kg per olive tree at the end of winter, using urea, ammonium sulphate, or ammonium nitrate, mainly depending on soil properties [17,18]. Despite the economic importance of olive tree cultivation in the Mediterranean region, there is not enough information regarding olive nutrient requirements, mainly nitrogen [19]. Fernández-Escobar et al. [20] concluded that the application of fertilizers does not have a significant impact on crop yield, fruit weight, oil content, or vegetative growth after a period of five years. Ferreira et al. [21] argue that previous studies on nitrogen fertilization in olive groves, while important, lack conclusive guidance for fertilizer recommendation systems. Consequently, this topic holds scientific interest and carries substantial practical significance within the field of olive grove fertilization.

The olive tree holds an important position in the Albanian economy as one of the main fruit trees. The olive cultivar "Kalinioti" is the dominant variety in Albania, covering more than 55% of the total olive tree area [22]. This is attributed to its remarkable adaptability to a range of environmental conditions and its high oil content. The lack of data regarding nitrogen fertilization of the olive variety "Kalinioti", in conjunction with the fact that olive fertilization is usually based on visual practices, has resulted in the thoughtless and excessive use of nitrogenous fertilizers that many times adversely affect productivity, fruit quality, olive oil production, and groundwater quality [23-25]. Moreover, basal fertilization with K and P has become an established and necessary cultivation practice in olive orchards in the region of South Albania, aiming at a balanced fertilization of nitrogen (N), phosphorus (P_2O_5), and potassium (K_2O). Potassium is considered important for fruit production [26,27]. In addition, due to the fact that phosphorus fertilizers are rapidly fixed to the soil, particularly one rich in $CaCO_3$, a large proportion of applied P may become chemically bound, whereas only a small fraction of soil P remains in the soil solution and is available for plant uptake [28,29], restricting reproductive growth and yields [28]. Therefore, olive leaf nutrient content (N, P, and K) is a valuable tool to determine the NPK status of olive trees. The aim of this work was to study the effect of nitrogen fertilization on the vegetative growth, yield, fruit set, fruit weight, and fruit oil concentration of the olive cultivar "Kalinioti" and its leaf nutrient (N, P, K) content. This study was based on 6-year field trials in commercial orchards of mature trees where intensive olive cultivation was being practiced.

2. Results

A significant and positive impact of N fertilizer doses on shoot length was recorded (Table 1). Tree crown volume showed the same tendency with the amount of N applied. At the highest level of N, trees showed excessive vegetative growth with many water shoots. Nitrogen fertilization positively affected olive yield compared to control. The increase was significant with the application of 4 kg NH₄NO₃/tree (~265 kg ha⁻¹), where yield

reached 62.5 kg/tree compared to 37.1 kg/tree of the control. Further increases in N rates significantly reduced yield while increasing the number of water shoots.

Table 1. The effect of ammonium nitrogen fertilizer on fruit yield, shoot length, and canopy volume of olive (cv. Kalinioti) trees.

Treatments ⁽²⁾	Yield (kg/tree)	Shoot Length (cm)	Canopy Volume (m ³)
Control	37.09 <i>d</i> ⁽¹⁾	9.2 <i>g</i>	18.2 <i>c</i>
1 kg NH ₄ NO ₃ /tree	39.2 <i>d</i>	11.4 <i>f</i>	19.4 <i>c</i>
2 kg NH ₄ NO ₃ /tree	45 <i>dc</i>	12.8 <i>e</i>	19.8 <i>bc</i>
3 kg NH ₄ NO ₃ /tree	54b	16.9 <i>d</i>	21.5 <i>b</i>
4 kg NH ₄ NO ₃ /tree	62.5 <i>a</i>	19.3 <i>c</i>	21.0 <i>bc</i>
5 kg NH ₄ NO ₃ /tree	49.0 <i>c</i>	24.7 <i>b</i>	24.7 <i>a</i>
6 kg NH ₄ NO ₃ /tree	46.4 <i>cd</i>	30.2 <i>a</i>	25.8 <i>a</i>
ANOVA table F value ⁽³⁾	13.54 **	5741.4 ***	101.86 ***

⁽¹⁾ Means (values of 6 years) in the same column followed by different letters denote significant differences according to Duncan's multiple range test (p < 0.05). ⁽²⁾ No. of samples per treatment and year (Yield: 15 trees; Shoot length: 120 shoots; Canopy volume: 15 trees). ⁽³⁾ Values of F: ** p < 0.01; *** p < 0.001.

As regards fruit set, a significant increase in the initial fruit set was recorded due to the application of 3 and 4 kg NH_4NO_3 /tree compared to the control (Table 2). Application of 4 kg fertilizer/tree gave the highest initial and final fruit sets (7.63% and 3.03% respectively) among treatments. A further increase in N fertilizer (5 kg NH_4NO_3 /tree and 6 kg NH_4NO_3 /tree) reduced fruit sets.

Table 2. The effect of ammonium nitrogen fertilizer on initial (May–June) and final (November) fruit set of olive (cv. Kalinioti) trees.

Treatments ⁽²⁾	Initial Fruit Set (May–June) (%)	Final Fruit Set (November) (%)
Control	3.07e ^{(1) (3)}	0.7 <i>d</i>
1 Kg NH ₄ NO ₃ /tree	3.55 <i>d</i>	1.13 <i>d</i>
2 Kg NH ₄ NO ₃ /tree	5.32 <i>c</i>	1.79 <i>c</i>
3 Kg NH ₄ N0 ₃ /tree	6.09 <i>b</i>	2.53 <i>b</i>
4 Kg NH ₄ N0 ₃ /tree	7.63 <i>a</i>	3.03 <i>a</i>
5 Kg NH ₄ N0 ₃ /tree	5.91 <i>b</i>	1.71 <i>c</i>
6 Kg NH ₄ N0 ₃ /tree	5.19 <i>c</i>	1.65 <i>c</i>
ANOVA table F value ⁽³⁾	1275.31 ***	184.877 ***

⁽¹⁾ Means (values of 6 years) in the same column followed by different letters denote significant differences according to Duncan's multiple range test (p < 0.05). ⁽²⁾ No. of selected flowers per treatment and year: 300. ⁽³⁾ Values of F: *** p < 0.001.

Fresh weight of 100 olives, oil content, and maturation index increased with time, reaching maximum levels at 5–10 December (Table 3). Although the trees that received 4 kg NH₄NO₃ had the maximum yield, the weight of 100 olives (W_{100}) in these trees was much lower compared to the other treatments. The application of nitrogenous fertilizer had no effect on oil content, but higher fertilizer applications (5 and 6 kg NH₄NO₃/tree) in December significantly reduced the maturation index.

	1–5 November		20–25 November		5–10 December			20–25 December				
	W ₁₀₀	OC	M.I.	W ₁₀₀	OC	M.I.	W ₁₀₀	OC	M.I.	W ₁₀₀	OC	M.I.
Treatments ⁽²⁾	g	%	%	g	%	%	g	%	%	g	%	%
Control	336b ⁽¹⁾	21.8	2.13 <i>a</i>	360a	24.2	3.05 <i>a</i>	389a	27.1	3.66 <i>a</i>	384 <i>a</i>	26.2	3.37 <i>a</i>
1 kg NH ₄ NO ₃ /tree	325b	21.3	2.10 <i>a</i>	353a	23.8	3.15 <i>a</i>	375a	26.8	3.75 <i>a</i>	375 <i>a</i>	26.5	3.44 <i>a</i>
2 kg NH ₄ NO ₃ /tree	321 <i>b</i>	21.2	2.18 <i>a</i>	342 <i>ab</i>	24.5	3.21 <i>a</i>	370a	27.2	3.82 <i>a</i>	369a	26.7	3.48 <i>a</i>
3 kg NH ₄ NO ₃ /tree	318b	21.0	2.12 <i>a</i>	329b	23.7	3.28 <i>a</i>	365 <i>ab</i>	27.5	3.78 <i>a</i>	361 <i>a</i>	26.0	3.51 <i>a</i>
4 kg NH ₄ NO ₃ /tree	381 <i>a</i>	20.9	2.10 <i>a</i>	309 <i>c</i>	23.5	3.15 <i>a</i>	337b	26.3	3.66 <i>a</i>	331 <i>b</i>	25.8	3.55 <i>a</i>
5 kg NH ₄ NO ₃ /tree	315c	20.8	1.95 <i>ab</i>	345 <i>ab</i>	24.1	2.75ab	374 <i>a</i>	26.8	3.16b	370 <i>a</i>	25.9	3.00 <i>b</i>
6 kg NH ₄ NO ₃ /tree	312 <i>c</i>	21.1 NS	1.87b	340 <i>ab</i>	23.7 NS	2 .71 <i>b</i>	378a	27.0 NS	3.11 <i>b</i>	373 <i>a</i>	26.1 NS	3.01 <i>b</i>
ANOVA table, F value ⁽³⁾	664.30 ***	1.99 NS	10.72 **	211.20 *	1.66 NS	43.12 ***	179.17 *	1.59 NS	39.14 **	202.18 ***	1.41 NS	35.05 **

Table 3. The effect of time collection, on weight of 100 olives (W₁₀₀, g./tree), oil content (OC, %), and maturation index (M.I., %) of olive (cv. Kalinioti) trees.

⁽¹⁾ Means (values of 6 years) in the same column followed by different letters denote significant differences according to Duncan's multiple range test (p < 0.05). ns: no significant differences. ⁽²⁾ No. of samples per treatment and year: W100:15; OC: 15; M.I.: Each of the 15 measurements is based on 100 olive fruits. ⁽³⁾ Values of F: * p < 0.05; ** p < 0.01; *** p < 0.001; NS: no significant differences.

With increasing N doses, leaf N concentration increased, reaching 2.38% at the level of 6 kg NO₃NH₄/tree (Table 4). Maximum yield (62.5 kg/tree at the dose of 4 kg/tree) corresponded to 2.01% N in leaves. Control trees and trees receiving 1 kg NO₃NH₄ had leaf N concentrations (1.23–1.34%, d.w.) well below the optimum levels (1.5–2.3%) [30,31] and close to the deficient threshold range (1.2–1.4%) [20,32–34]. High levels of fertilizers (5–6 kg NH₄NO₃/tree) reduced P and K concentrations in leaves compared to control; however, the decrease was significant only for K. Leaf P and K concentrations were within the optimal range (0.09–0.3% for P and 0.70–1.0% for K) [31,34,35], with the exception of the concentration of K in the leaves of trees receiving N dosages (5–6 kg NH₄NO₃/tree) (0.4%, d.w.), where values were far below the optimum range.

Table 4. The effect of ammonium nitrogen fertilizer on N, P, and K concentration of leaves of olive (cv. Kalinioti) trees.

Treatments ⁽²⁾	N (%, d.w.)	P (%, d.w.)	K (%, d.w.)	
Control	1.23e ⁽¹⁾	0.123	0.78 <i>a</i>	
1 kg NH ₄ NO ₃ /tree	1.34 <i>e</i>	0.125	0.75 <i>a</i>	
2 kg NH ₄ NO ₃ /tree	1.55 <i>d</i>	0.128	0.77 <i>a</i>	
3 kg NH ₄ NO ₃ /tree	1.89c	0.131	0.73 <i>a</i>	
4 kg NH ₄ NO ₃ /tree	2.01 <i>b</i>	0.129	0.69 <i>ab</i>	
5 kg NH ₄ NO ₃ /tree	2.29 <i>a</i>	0.120	0.61 <i>b</i>	
6 kg NH ₄ NO ₃ /tree	2.38a	0.115 NS	0.57 <i>b</i>	
ANOVA table, F value ⁽³⁾	401.4 ***	1.892 NS	16.812 ***	

⁽¹⁾ Means (values of 6 years) in the same column followed by different letters denote significant differences according to Duncan's multiple range test (p < 0.05). ⁽²⁾ No. Samples per treatment and year: 15. ⁽³⁾ Values of F, *** p < 0.001; NS: no significant differences.

3. Discussion

Excess nitrogenous fertilization significantly enhanced vegetative growth. The canopy volume of trees receiving 5 and 6 NH_4NO_3 /tree was significantly higher compared to that of trees treated with lower N rates; however, overfertilization did not enhance yield. Indeed, further increases in N rates (>4 kg fertilizer or above 265 kg N/tree) reduced yield and increased the number of water shoots since an abundant supply of nitrogen, sufficient water, high air humidity, and moderate temperatures stimulate vegetative growth [36]. Similarly, Haberman et al. [5] reported that the highest N level was the most effective in promoting vegetative growth but did not induce an increase in yield, whereas low nitrogen fertilization has negative effects on both vegetative growth and the production of olive fruit and oil content. These effects were attributed to a decrease in the intensity of flowering and a reduction in the rate of perfect flowers and fruit set. Concentration of N can be correctly detected through a leaf analysis, which is the best diagnosis method to determine the nutrient status and plan fertilizer recommendations [37]. In our study, leaf N concentrations of 2.29–2.38% in trees treated with 5–6 kg of NH_4NO_3 were associated with reduced yields. However, even control trees showed leaf P and K concentrations within the optimum range, which is probably due to the fact that basic soil analysis showed an excessive concentration of P (120 mg kg⁻¹) and an adequate level of available K (0.45 meq/100 g). The relatively higher yields achieved at 3 and 4 kg of fertilizer per tree (about 204 and 265 kg N ha^{-1} respectively) in this study seem to be necessary for a satisfactory production. Under these N doses, the concentration in leaves ranged from 1.89–2.01% d.w., within optimum levels for olive trees. These N doses are higher compared to nitrogen doses $(40-140 \text{ kg N ha}^{-1})$, which are characterized as the best for both yield [38,39], and guarantee the quality and stability of the olive oil. In fact, annual applications of $80-200 \text{ kg N} \text{ ha}^{-1}$ and more are common in many areas of the Mediterranean basin [1]. Variations in soil climatic conditions, different cultivars, phenological stages, cultivation areas, etc. are the causes of the different responses of olive orchards to various N fertilization rates [40,41].

Olive trees in southern Spain's arid regions were fertilized annually with varying quantities of nitrogen. Fernández-Escobar and Marín [17] and Fernández-Escobar et al. [32] concluded that after three and six years of experimentation, respectively, increasing the amount of N applied from 0 to 1 kg of N per olive tree (about 2.86 kg NH₄NO₃/tree) did not result in an increase in yield, fruit size, oil content, or vegetative growth. Fernández-Escobar and Marín [17] suggested that there is no need for annual application of nitrogen fertilizer in olive orchards in order to achieve satisfactory productivity and growth, as long as the leaf nitrogen levels remain above the sufficiency threshold. Ângelo Rodrigues et al. [42] recommend adjustments to the rates of N every year to prevent reductions in tree crop performance and improve nutrient-use efficiency.

With regards to oil content, our results showed that nitrogen fertilization did not significantly affect it. This may be attributed to the fact that soil nitrogen content levels under control conditions are characterized as medium in total N (1.47 mg/g), and therefore nitrogen enrichment might only affect oil production under conditions of low nitrogen availability. Fernández-Escobar et al. [32] also reported similar findings, where increasing the amount of nitrogen applied from 0 to 1 kg of N per olive tree did not result in an increase in oil content. In addition, Fernández-Escobar and Marin [17] observed lower oil content in trees with leaf nitrogen concentrations below the deficient threshold of 1.4%, suggesting a delay in lipogenesis. In our study, leaf nitrogen concentration under control treatment was below the deficient threshold, but oil content remained unaffected by increasing N doses. In contrast, other authors have shown that nitrogen fertilization has positive effects on oil content [43]. Erel et al. [44] highlighted the significance of balanced N nutrition in oil olive cultivation for optimizing oil content production. A number of researchers have stated that limited nitrogen availability in the soil and excessive nitrogen fertilizer have a negative impact on flowering and fruit set [13,36,42,45]. In our study, the final fruit set was significantly enhanced in trees treated with 3-4 kg NH₄NO₃/tree compared to the rest of the N doses, indicating that improving fruit set may improve yields. On the other hand, several studies show that when the number of flowers is artificially reduced, the fruit set increases proportionally, resulting in a similar fruit load [46,47]. Moreover, leaf N concentrations under control and $1 \text{ kg NH}_4\text{NO}_3$ /tree were below the optimum range (1.5–2.3%), indicating limited soil N availability, which leads to a reduction in the size of the photosynthetic apparatus and its efficiency [38]. According to [48], N deficiency has an adverse impact on fruit set, probably due to inadequate total quantities of nutrients available to the flowers when they develop into fruit. In addition, Fernández-Escobar et al. [8] noted that N in excess has a similar negative effect on flowers as N deficiency does. Erel et al. [13] did an experiment in a container to see how different amounts of nitrogen, phosphorus, and potassium in the irrigation solution affected the flowering and fruit set of an olive tree (Olea europaea L. cv. Barnea). They observed that the highest N concentration in the irrigation solution (14.1 mM) decreased fruit set, but to a lesser extent than flowering.

Table 4 shows that the fruit weight, oil content, and maturation index increased over time, reaching their maximum values on 5–10 December, while no further increase was recorded during the following olive harvesting period from 20–25 December. This indicates that the appropriate time period to start olive harvesting is 5–10 December in order to avoid early cropping, which can reduce oil yields, or to avoid yield losses due to fruit shedding derived from late harvests. The addition of 4 kg NH₄NO₃/tree resulted in maximum yield, but the weight of 100 olives was significantly lower compared to the rest of the fertilization rates, which is not desirable for table olives. According to Silva et al. [38], nitrogen application decreased the fruit's weight and size due to photosynthate partitioning. In addition, elevated nitrogen applications (4–6 kg NH₄NO₃/tree) had a negative impact on the maturation index relative to the control by extending the colouration period and fruit maturation [12,13,43]. The effect was more pronounced in trees receiving the two highest N doses (5–6 kg NH₄NO₃/tree). Silva et al. [38] reported that nitrogen application prolonged the period of colouration, reducing the fruit maturation index, particularly when applied at high rates (120 N kg ha⁻¹). Fernández-Escobar et al. [49] observed a delay in fruit maturation in trees with elevated nitrogen levels. This delay was attributed to the elevated concentration of anthocyanin, a pigment responsible for the black colour of olive fruits.

4. Materials and Methods

Twenty-five-year-old irrigated olive trees of variety "Kalinioti" that had been grafted onto wild olive rootstock (O.e.v. Olester) were selected for the field trials. The experiment was conducted for 6 years at the Pomologie Research Centre in Agioi Saranta, South Albania. The trees were planted with a spacing of 7 m by 7 m, resulting in a density of approximately 204 trees per hectare. In addition, every year, the trees received a light pruning regime. Weeds were chemically controlled by applying glyphosate (N-phosphonomethyl) glycine at a rate of 3 L/ha. We applied the glyphosate to the vegetation during the early spring season. There were no additional phytosanitary products used throughout the duration of the experiment. The area has a hot-summer Mediterranean climate (Csa) as of the Köppen climate classification. Monthly mean temperature (maximum, average, and minimum) and rainfall are shown in Table 5. During the years of experimentation, the area experienced a mean annual precipitation of about 781 mm, with the minimum temperature (2.8 °C) recorded in January and the highest temperature (34.4 °C) recorded in July. The soil of the experimental field was clay loamy, with slightly alkaline pH 7.8, rich in total $CaCO_3$ (10.3%), poor in organic matter (1.2%), medium in total N (1.47 mg/g), relatively high in available K (0.45 meq/100 g) and very rich in available phosphorus (120 mg kg⁻¹), indicating that phosphorus is more likely to be lost by runoff since levels of P-Olsen exceeded 60 mg kg^{-1} [50]. The experimental design included seven treatments, corresponding to seven ammonium nitrate (50% NH4+, 50% NO3) rates, 0 (control) to 6 kg/tree, which corresponded to approximately 0, 0.3, 0.7, 1.0, 1.4, 1.7, and 2 kg N/tree, respectively, or to 0, 61, 143, 204, 265, 347, and 408 kg ha⁻¹ of N, respectively. These fertilization regimes were applied to three groups (three replicates) of five equally sized and well-developed olive trees. The selection of trees was based on criteria of uniformity for vigour and fruit load. Fertilizer was applied on a per-tree basis. Half of the fertilizer was given at the beginning of the annual cycle, in the first 10 days of March, and the rest was given at the fruit set [26], which approximately corresponded to the end of May and the beginning of June. Nitrogen fertilizers were distributed evenly on the soil over the green cover without burying. In addition to the N fertilizer treatments in the experimental design, all trees received a basal fertilizer application plan. In particular, 160 kg ha⁻¹ of K_2O as potassium sulphate and 56 kg ha⁻¹ of P₂O₅ as superphosphate were incorporated into the soil at the end of February, according to farmer cultivation practices. Trees were drip-irrigated with one hundred litres of water per week.

tation, and relative numidity throughout the duration of the six-year trial period.													
Average Values	Jan.	Feb.	Mar.	Apr.	May.	Jun.	Jul.	Aug.	Sep.	Oct.	Nov.	Dec.	•
High Temp. (°C)	13.3	13.9	16.7	19.4	24.4	31.7	34.4	33.9	28.3	23.3	18.3	13.9	
Temp. (°C)	6.7	7.2	11.7	15.0	20.6	24.4	27.2	28.3	21.7	18.3	12.2	8.3	

16.7

16.5

52

Table 5. Average monthly temperature (including both the minimum and maximum values), precipitation, and relative humidity throughout the duration of the six-year trial period.

20.0

8.1

51

19.4

21.3

56

16.7

61.0

66

11.7

96.5

73

7.8

132.1

75

4.4

119.4

76

4.1. Measurements

5.6

70.6

75

Low Temp. (°C)

precipitation (mm)

Relative humidity (%)

2.8

87.6

76

3.3

89.4

74

9.4

47.5

75

13.9

31.2

65

1. Vegetative growth: The length of 20 tagged shoots located at the four points of the horizon was recorded every year in two trees from each group (replicate), for a total of 40 shoots for each group.

2. Fruit set: The fruit set was expressed as the percentage of the initial number of flowers resulting in viable fruit. One hundred contiguous flowers on the four individual branches from the middle of the crown of one tree/replicate, located at the four points of the horizon, were labelled during the flowering period. These branches were used to determine the initial fruit set at the end of May, the beginning of June, and the final fruit set in November, which was actually the percentage of olive fruits remaining on the tree.

3. Olive yield: The harvest took place in the second half of December. The harvest was performed using wooden sticks, rakes, and small portable machines with spinning rubber 'fingers' that shake the olives from the branches. Five trees were harvested from each group and treated. The olive yields (kg/tree) were weighed separately per labelled tree.

4. Fruit weight (W_{100}): The fresh weight of 100 fruits was measured from each tree in the groups (replicates), and thus a total of 500 fruits/group was taken. The fruits were weighed separately for each tree and group, and the weight of 100 fruits was calculated. Samples were taken every 15 days during November and December.

5. Maturation index (M.I.): Samples of 100 olive fruits per selected tree in each group (replicate) were selected every 15 days during November and December and were categorized into 8 colour classes based on epidermis and pulp colour (0 to 7) in order to determine the maturation index according to the following equation [51].

M.I. =
$$(\alpha \times 0 + b \times 1 + c \times 2 + d \times 3 + e \times 4 + f \times 5 + g \times 6 + h \times 7)/100$$

where a–h, are the numbers of fruits in each category and 1–7 are the numbers of colour classes.

The olives were classified into the following categories: 0, olives with intense green epidermis; 1, olives with yellowish-green epidermis; 2, olives with red spots or areas in less than half of the fruit; 3, olives with red or light violet epidermis over more than half; 4, for black epidermis and white pulp; 5 if the epidermis is black and less than half pulp is purple; 6, if the epidermis is black and more than half pulp purple (without reaching the stone); 7, if the epidermis is black and total pulp purple (reaching the stone).

Olive fruit samples were the same as those used the determination of the fruit weight (W_{100}) .

6. Canopy volume: For the determination of the canopy volume the following equation was used according to Roose et al. [52]:

 $V = 4/6 \times \pi \times H \times R^2$ where V= volume (m³), $\pi = 3.14$, H = tree height (m) and R = radius of canopy

The canopy volume was measured separately per labelled tree.

7. Fruit chemical analysis: Oil content was determined on a sample of about 100 fruits per selected tree (5 trees per group), without any kind of infection or physical damage, by extracting dry material with petroleum ether at 40–60 °C using a Soxhlet apparatus. Olives were dried at 70 °C in a ventilated oven until a constant weight was measured in two successive weighing measurements. Then, olives were ground in a mortar and the paste was weighed and analysed by the Soxhlet apparatus [53].

8. Leaf analysis: Samples of 40 fully-expanded, mature leaves (5–8 months old) were collected from each of the five trees in each group at the end of July and from the middle portion of non-bearing, current-season shoots, taken at shoulder height, according to the method described by Koukoulakis and Papadopoulos [54]. Leaf samples were taken after a minimum of 6–8 weeks following full bloom, which typically occurs in mid-spring around April/May. This period was chosen because it guarantees that the nutrient levels in the leaves have reached a more stable state. Once in the laboratory, samples were washed using 1% Triton X-100 (Merck, Rahway, NJ, USA), and rinsed three times with water and deionized water. Moisture was eliminated using filter paper and the samples were dried in paper envelopes at 70 °C until they reached a constant weight. Dried samples were ground in a stainless "Fritch" pulverisette mill to pass through the 1 mm round whole sieve. Ground samples were stored at room temperature in acid-washed glass jars. Leaf

samples were analysed for total N, P, and K. Total N was determined by the Kjeldahl method, using a semiautomatic analyser Buchi, B324 (Büchi, Flawil, Switzerland). Prior to the measurement of the other nutrients, leaf subsamples were subjected to dry ashing at 520 °C for 6 h, then diluted with hydrochloric acid (HCl) in a 1:1 ratio v/v [55]. Phosphorus was determined calorimetrically in the same solution by the vanado-molybdo-phosphoric method [56] using an UV-Vis spectrophotometer. Total K was determined through atomic absorption spectrophotometry (PerkinElmer Analyst 100 atomic absorption spectrometer, PerkinElmer, Inc., Waltham, MA, USA).

4.2. Statistical Analysis

The evaluation of the effect of the fertilizer treatments in each year of experimentation was provided by one-way ANOVA using SPSS version 21. Before performing analysis of variance, the Shapiro–Wilk ($p \le 0.05$) and Bartlett ($p \le 0.05$) tests were applied to test normality and homogeneity of variances, respectively. Duncan's multiple-range test was used to test differences among means (p < 0.05).

5. Conclusions

In conclusion, considering the current experimental conditions, the application of $4 \text{ kg NH}_4\text{NO}_3/\text{tree}$ (265 kg N ha⁻¹) gave the highest yield while significantly reducing the weight of 100 olives, and therefore this dose is recommended for olive oil production. Similar yields were also achieved by the lower dose of fertilizer, 3 kg NH₄NO₃/tree (204 kg N ha⁻¹), without reducing olive weight, indicating the appropriate fertilizer dose for table olives. Further additions of fertilizer caused reduced growth and a delay in fruit maturation. The best period for harvesting olives cv. Kalinioti seemed to be the beginning of December (5–10) when olive weight, oil content, and maturation index were at their maximum levels.

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