THE EFFECT OF COMMERCIAL SEAWEED EXTRACTS AND COMMERCIAL LIQUID ORGANIC NITROGEN FOLIAR SPRAYS ON PRODUCTIVITY, OIL QUALITY AND NUTRITIONAL STATUS OF THE OLIVE CULTIVAR ‘MASTOIDIS’

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Abstract

Commercial products so-called biostimulators and liquid organic nitrogen fertilization treatments were applied foliarly in ‘Mastoides’ olive cultivar in order to study their effects on productivity, olive oil content, olive oil quality and the nutritional status of the trees based on the concentrations of the basic inorganic elements in leaves. The treatments were the following: a) control, foliar application of b) of Seamac-PCT™ (0.5% v/v, of the seaweed *Ascophyllum nodosum*), c) Acadian (0.5% w/v of the seaweed *Ascophyllum nodosum*) and d) the commercial product N-organics (0.5% v/v). Foliar sprays were applied once at the termination of the flowering period, whereas no other fertilization treatment was applied foliarly or from the soil. Acadian increased significantly olive tree productivity, whereas Seamac-PCT™ increased significantly oil content compared to the rest fertilization treatments. However, total phenol concentration in oils was minimized significantly in Acadian and Seamac-PCT™ fertilization treatments whereas, the application of N-organics led to a significant increase in their concentration compared to control. Phenolic compounds affect the taste, in particular the positive bitterness organoleptic attribute, the oxidative stability of virgin olive oil and have beneficial effects on human health. The olive oil in all fertilization treatments, including the control, was qualified as ‘extra virgin’ based on the measured free acidity, the spectroscopic indices K_{232} and K_{270} and the peroxide index. The application of Seamac-PTC™ increased significantly K, Mn, B and Cu and concentration in leaves, the application of Acadian increased the concentration of P, while both Seamac-PCT™ and Acadian increased significantly the concentration of Zn. Moreover, both Acadian and N-organics increased significantly leaf Mg concentration. Iron and N were significantly increased in all treatments compared to the control.

Keywords: *Ascophyllum nodosum*, fertilization, olive oil quality, organic nitrogen, nutrition

Introduction

One of the factors that exert great influence on olive tree growth and fruit quality is the fertilization. Seaweed extracts, being organic and biodegradable, are important in sustainable agriculture. Plants grown in soils treated with seaweed manures, or extracts applied either to the soil or foliage, exhibit a wide range of responses, such as increased crop yield, quality and uptake of inorganic elements from the soil, resistance of plants to stress conditions, reduced incidence of fungal and insect attack and lower productivity cost that have been well documented in a number of cultivations (Crouch and Staden, 1992, Chouliaras et al., 1997, Fornes et al., 2002). A significant improvement in the size of olives and the quality of olive oil was recorded in trees sprayed with an *A. nodosum* extract fortified with added nitrogen and boron (Chouliaras et al., 2009). However, there is not adequate information in the literature about the effects of seaweed extracts applied foliarly on olive tree yield, fruit growth and oil quality. However, foliar application of commercial products based on organic nitrogen, instead of inorganic nitrogen fertilizers have been used during the last two decades in Greece, for the production of olives under sustainable agriculture conditions. Nitrogen exerts a significant effect on olive plant growth, reduces biennial bearing and increases the percentage of perfect flowers and yield (Fernandez- Escobar et al, 2004).

Olive oil quality is based directly on free acidity, the spectroscopic indices K_{232} and K_{270} and the peroxide index (EC, 1991) and indirectly on the content of total and individual phenolic compounds. Phenolic compounds affect the taste being positively correlated with bitterness organoleptic attribute, they influence the oxidative stability of virgin olive oil and they have beneficial effects on human health related to the prevention of cardiovascular diseases, cancer and neuro-degenerative diseases (Shahidi, 1996). Despite these positive aspects, consumers prefer virgin olive oils with low or moderate levels of bitterness.

The aim of the current research was to study the effects of commercial products derived from the seaweed *Ascophyllum nodosum* (Seamac-PCT^{TM}, Acadian) and the commercial product N-Organics on productivity, fruit growth, leaf nutrient status, olive oil content and quality of the Greek olive cultivar ‘Mastoidis’.

Materials and methods

The experiment was carried out in an orchard of 15-year old olive trees of the cv. ‘Mastoidis' or 'Tsounati' (locally called 'Athinoelia'), placed at Neapoli Lakonias (Peloponnesus, Greece). Four fertilization treatments were applied on five trees-replications, 2.5 m height, planted at distances of 7.5 m. Olives were picked at the same time (stage of full maturity), during the harvesting period 2010-2011, seven months after the termination of the flowering period. During this study, the minimum, maximum and mean temperatures were 7.4^{o}C, 35.2^{o}C and 26.7^{o}C, respectively, and the mean annual rainfall was 883 mm. Olive trees were sprayed with Cu (0.05% w/v) against fungal diseases during early April and September, whereas McPhail traps were used against the olive fly (one trap per tree).

The treatments applied were the following: a) control, b) foliar application Seamac-PCT^{TM} (0.5% v/v of the seaweed *Ascophyllum nodosum*), c) foliar application of Acadian (0.5% w/v of the seaweed *Ascophyllum nodosum*) and d) foliar application of the commercial product N-Organics (0.5% v/v). Foliar sprays were applied once at the termination of the flowering period, whereas no other fertilization treatment was applied foliarly or from the soil.

The olive fruits (75 kg of each treatment) were collected the same period of time at the optimum maturity stage and were processed into a three-phase decanter using the same protocol for all samples. Olive oil was finally filtered and stored in dark green bottles of 0.5L capacity, at 4°C, until analysis. Three olive oil samples from each treatment were analysed.

2.1.1 Quantitative papameters

Olive productivity was recorded as kg per tree from each replication, whereas as mean fruit weight was recorded the mean of 15 fruits from each-tree replication.

Olive oil content was estimated with a Soxhlet apparatus, using 10 fruits of representative size from each one of the five tree-replications pre treatment.

The quiantity of olive oil per tree was estimated by multiplying the yield (Kg of olives per tree with the proportional percentage of olive oil.

2.2 Oil quality indices

Free acidity, expressed as percentage of oleic acid, peroxide value (PV), expressed as milliequivalents of active oxygen per kilogram of oil (meq O_2/kg), and K_{232} and K_{270} extinction coefficients, calculated from absorption at 232 and 270 nm, were measured using the analytical methods described in European Commission Regulation EEC 2568/91 (EC, 1991). Solvents were purchased from Merck (Darmstadt, Germany).

2.3 Phenolic compounds

An aliquot of the aqueous- methanolic solution of phenolic compounds extracted from virgin olive oil. Total phenols were evaluated colorimetrically at 725 nm with the Folin-Ciocalteau reagent (Gutfinger, 1981) in an Ultraspec II (LBK Biochrom, Cambridge, England). The calibration curve is constructed using standard solutions of gallic acid within the range of 0.01 to 1 mg/mL.

2.4. Leaf nutrient analysis

Leaves from the middle of non-fruiting shoots were collected in late October, washed with a 0.01% soap solution and 0.1 N HCl and rinsed twice with distilled water. After drying at 68^oC for three days, the samples were ground to a fine powder and 0.5 g was dry ashed at 500^oC in a muffle furnace for 5 h. The ash was diluted in HCl 6N and analysed for K, Ca, Mg, Fe, Mn, Zn and Cu by atomic absorption spectrophotometry. Phosphorus concentration was determined spectrophotometrically with the phosphovanado-molybdate method, N with the Kjeldahl method and B with the azomethine-H method.

The statistical analysis was performed according to the complete block randomized design (five treatments and five replications per treatment), while analysis of variance and Duncan’s multiple range test were used to compare differences between means.

Results and Discussion

According to the results of the current research, the application of N-organics caused the highest fruit growth among all the rest treatments and the highest productivity when combined with Seamac-PTC^{TM} (Table 1). Seamac-PTC^{TM} is a commercial product based on the sea algae *Ascophyllum nodosum*. *Ascophyllum nodosum* is rich in cytokinines, auxins, gibberellins, minerals, betaines, aminoacids, proteins and oligosaccharides (Cardozo, 2007, Khan et al., 2009, Craigie, 2011). According to the above researchers, this extract influences cell growth and division cycle, expansion, nutrition and maturity as well.

Organic nitrogen fertilization has been achieved high yield in maize and barley, ensured high increases of total organic carbon and showed the least N mineral soil deficit (Montemurro et al., 2006). The high amount of aminoacids which is contained in the commercial product containing N-organics could be the reason for the highest increment in olive fruit size. According to Ashmead (1986), aminoacids improve the original cell ultrastructure resulting in higher photosynthetic efficiency. Also, it is suggested that they stimulate the activity of some enzymes responsible for protein and carbohydrate synthesis and therefore the biomass that is directly correlated with the yield. Olives treated with Seamac-PTC^{TM} gave by far the highest olive oil content and finally the highest production of olive oil per tree. Acadian and N-organics presented almost similar results, concerning the production of olive oil per tree which was proved to be twice compared to control (Table 1). Similar results about the effects of SWE on fruit growth and maturation have been reported for other fruit species such as kiwifruit (Chouliaras et al., 1997). Furthermore, similar effects concerning fruit size and maturity have been reported for 'Clementine' mandarin and 'Navelina' orange trees sprayed with an *A. nodosum* product (Fornes et al., 2002).

Table 1. The effect of fertilization treatments on mean fruit weight, yield, olive oil content and olive oil quantity of the olive cv. 'Mastoidis'

Treatment	Fruit weight (g)	Yield (kg/tree)	Oil content (%)	Olive oil (kg/tree)
Control	2.75 c	15.7 c	14.8 c	2.32 c
Seamac-PCT^{TM}	2.85 bc	24.5 b	25.9 a	6.57 a
Acadian	3.13 b	31.4 a	15.8 c	4.96 b
N-Organics	3.58 a	25.2 b	18.6 b	4.72 b

The statistical analysis was performed by Duncan's test (*P*=0.05). Means in the same column with different letters are statistically different.

The results for acid value, extinction coefficients, K_{232}, K_{270} and peroxide values are within the limits of European Community Regulations (EC, 1991), implying that the oils sampled from all treatments, including the control, are classified as 'extra virgin' (Table 2).

Table 2. The effect of fertilization treatments on quality and physicochemical characteristics of olive oil of the cv. 'Mastoidis'

Treatment	Control	Seamac-PCT^{TM}	Acadian	N-Organics
Acidity (% oleic acid)	0.18±0.04	0.17±0.02	0.19±0.03	0.15±0.03
Peroxide value (meq kg^{-1})	5.83±0.35	6.48±0.64	8.45±0.38	6.34±0.19
K_{232}	1.58±0.04	1.53±0.03	1.53±0.03	1.64±0.04
K_{270}	0.1±0.01	0.1±0.01	0.11±0.01	0.11±0.01
Total phenols (mg kg^{-1} as gallic acid equivalents)	72,23±9,02 a	41,34±4,95 b	42,67±1,61 b	100,92±10,65 c

The statistical analysis was performed by Duncan's test (*P*=0.05). Means in the same row with different letters are statistically different.

Total phenol concentration was significantly minimized in Acadian and Seamac-PCT™ fertilization treatments, whereas the application of N-organics led to a significant increase in their concentration compared to the control. The olive cultivar and the ripening stage of the fruit, the malaxation conditions and the extraction system used to separate oil from the paste have been the most studied agronomic aspects that affect phenolic concentration in virgin olive oil (Servili and Montedoro, 2002). There is not information in the bibliography concerning the influence of nitrogen fertilization on the concentration of total phenols in olive oil. A significant improvement in the quality of olive oil and minimization of total phenols concentration was recorded in trees sprayed with an *A. nodosum* extract additionally to nitrogen and boron fertilization (Chouliaras et al., 2009). Thus, fertilization treatment seems to be of great importance for minor constituents' concentration.

Table 3 presents the effects of fertilization treatments on nutrient leaf concentration. The application of Seamac-PCT™ increased significantly K, Mn, B and Cu and concentration in leaves, the application of Acadian increased the concentration of P, while both Seamac-PCT™ and Acadian increased significantly the concentration of Zn. Moreover, both Acadian and N-organics increased significantly leaf Mg concentration. Iron and N were significantly increased in all treatments compared to the control. Similarly, SWE application additionally to N and/or B fertilization increased N, K, B, Mn and Cu concentration in 'Koroneiki' olive leaves (Chouliaras et al., 2009).

This experiment also shows that olive productivity is significantly correlated with leaf nutrient contents. Similar conclusion has been also referred for olive trees cv. 'Chondrolia Chalkidikis' cultivated in northern Greece (Almaliotis et al., 2005).

Table 3. The effects of fertilization treatments on the concentration of the macrinutrients N, P, K, Ca and Mg (%) and the micronutrients Fe, Zn, Mn, B and Cu (μg/g) in leaves of the cv 'Mastoidis'

Treatment	Control	Seamac-PCT™	Acadian	N-Organics
N	1.79 b	2.44 a	2.23 a	2.15 a
P	0.090 b	0.092 b	0.110 a	0.093 b
K	0.76 c	0.87a	0.81 b	0.79 bc
Ca	2.23 a	1.94 b	2.26 a	1.98 b
Mg	0.252 b	0.255 b	0.294 a	0.289 a
Fe	61.9 b	79.8 a	75.1 a	80.2 a
Zn	21.5 c	26.5 a	27.5 a	24.2 b
Mn	27.1 b	36.2 a	29.6 b	29.2 b
B	15.2 c	24.6 a	19.9 b	17.9 bc
Cu	3.8 d	6.2 a	5.5 b	4.9 c

The statistical analysis was performed by Duncan's test (P=0.05). Means in the same row with different letters are statistically different.

In conclusion, the application of commercial products derived from the *Ascophyllum nodosum*, either based on organic nitrogen influenced positively olive tree productivity and the olive oil quantity, maintained the olive oil quality and improved the nutritional status of the olive cv. ‘Mastoidis’.

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