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Use Efficiency of Cyanobacteria and Olive Vegetation Water (Cyano/Ovw) Biofertilizer for Olive Trees under Different Mineral Npk Levels

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THIS STUDY was conducted during the two growing seasons of 2013 and 2014 on 13-year old *Manzanillo* olive trees cultivated at 5×8 m apart in sandy loam soil under standard cultural practices and drip irrigation in a private farm located at km 50 of Cairo-Alexandria road in the north west of Egypt to evaluate the effect of the bio-organic fertilizer Cyano/OVW, formulated from a successful cultivation of cyanobacteria mixed strains (*Nostoc muscorum*, *Anabaena oryzae* and *Spirulina platensis*) in 1:1 tap water-diluted of olive vegetation water (OVW), on some soil properties, growth parameters, mineral content, flowering, fruit set, yield and fruit quality. The bio-organic fertilizer (Cyano/OVW) was applied as soil drench or as foliar spray application methods. This study also highlighted the economic feasibility of these practices in improving fruit quality and crop yields. As soil drench application, Cyano/OVW was diluted with tap water (1:1) and added at the rates of 24, 36 and 48 liter/tree combined with three levels of 100, 75 and 50% of the recommended mineral NPK fertilizers. As for foliar spray application, constant volume of Cyano/OVW (1 liter/tree) was diluted with tap water (1:5) and sprayed from 4 directions (100%), 3 directions (75%) and 2 directions (50%) surrounding the trees which all received 100% of the recommended mineral NPK fertilizers. The control received the full dose of the recommended mineral NPK fertilizers only. Results revealed that all tested parameters i.e., soil biological and chemical properties, vegetative growth, fruit quality, fruit yield, fruit oil content, some physiological parameters and mineral contents of leaves were improved while, total phenolic compounds in soil and leaves were reduced in response to Cyano/OVW bio-organic fertilizer soil drench or foliar spray applications methods comparing with control. Application of foliar spray of 2 directions (50%) surrounding the trees + 100% of the recommended mineral fertilizers during the six months from January to June, twice each month was the most promising comparing with other treatments in improving soil moisture, growth, quality and productivity of *Manzanillo* olive trees fruits. This treatment would be recommended under this study and also under similar conditions as confirmed by the economic study.

Keywords: Cyanobacteria, Olive Vegetation Water, Soil Drench, Foliar Spray.

Olive (*Olea europaea* L.) oil producing countries are annually facing a major environmental problem resulting from oil waste water, designated "olive mill wastewater" (OMW) Known as alpechin, which is generated in large quantities in a short period of time. These type of effluents is general characterized by very high organic load, due to high levels of phenolic compounds and sugars, and have minimum levels of nitrogen compounds and low pH and is therefore one of the most serious environmental problems in those countries (Hachicha *et al.*, 2009). Nevertheless, the use of OVW for agricultural purpose could provide a lower cost source for water and nutrients (Piotrowska *et al.*, 2011) particularly in arid regions, like Egypt, which suffering from serious water and soil organic matter deficiencies (Hachicha *et al.*, 2009 and Magdich *et al.*, 2012). It can be used as a soil conditioner/ fertilizers amendment and proposed as one of the most suitable methods to restore soil fertility and resolve the problem of their disposal (Abu-Zreig and Al-Widyan, 2002). Even though the direct application of olive mill wastes on soil can be an approach of recycling nutrients and organic matter, the continuous application of OVW to soil without any treatment might cause unfavorable impact on plants, soil microbial population and activity because of phenolic, fatty acid and mineral salts contents and high COD and BOD (Kavdir and Killi, 2008). Phenolics are a group of dangerous toxic organic pollutants that are toxic to all the living organisms even at lower concentrations (Pimentel *et al.*, 2008). Phenolic compounds at higher concentrations are very difficult to remove from the environment even by using physical and chemical techniques (Araña *et al.*, 2001). Micro-Algae have already been found to detoxify OVW (Duarte *et al.*, 2011). In addition, EI-Sheekh *et al.* (2012) explored the ability of cyanobacteria to degrade phenolic compounds either by reduction, oxidation or by induction of some enzymes that degrade these toxic compounds. Cyanobacteria, as biofertilizer, are known to possess the ability to form associations with vascular/non-vascular plants and produce growth-promoting substances (Nanjappan *et al.*, 2007). They also known to increase soil fertility by enhancing the available N and P levels and exhibited an economical view that it can compensate about 50% of the recommended doses of N, P and K (Mahmoud *et al.*, 2015). Thus, this present work intended to ensure the mitigation of chemical fertilizers usage in the field of olive cultivation and to reduce their potential adverse impacts on the environment and the economy. Therefore, the objectives of this study were to examine the effect of replacing mineral fertilizers partially by using Cyano/OVW biofertilizer and to investigate the best feasible application method in raising crop of *Manzanillo* olive trees and improving soil fertility in the sustainable agriculture system.

Materials and Methods

Olive mill wastewater

Olive mill wastewater (OVW) was obtained from FIFA farm (km 50 of Cairo-Alexandria, Egypt). The raw OVW samples (Table 1) were generated by the three-phase olive-oil extraction process for 2013 and 2014 seasons.

Cyano/OVW biofertilizer

Three cyanobacteria strains (*Nostoc muscorum*, *Anabaena oryzae* and *Spirulina platensis*) were kindly supplied from Department of Microbiology, Soils, Water and Environment Research Institute (SWERI), Agricultural Research Center (ARC), Giza-Egypt to be inoculated in OVW substrate medium. A mixed culture of the three strains was grown in tap water dilution of OVW (1:1 v/v) for two weeks under lab. conditions and continuous aeration. 50-liter plastic tanks were used to prepare Cyano-OVW biofertilizer for field experiments by diluting 20L of non-sterilized OVW with 20L tap water (1:1 v/v). Chemical constituents of OVW (Table 1) and Cyano/OVW biofertilizer (Table 3) were measured according to APHA (1998).

TABLE 1. Chemical analysis of OVW samples (Values are average of two seasons).

Parameters	**Vegetative water**
pH	5.01
EC	15.52
T.N %	1.4
Organic Matter %	20.81
Organic carbon %	12.1
C/N ratio	86.43
Total P %	0.73
Total K %	1.30
$N\text{-}NH_4$ (ppm)	25.50
$N\text{-}NO_3$ (ppm)	8.90
Total phenols (g/l)	9.46

Field experiment

Field practices

Two experiments were carried out on a 13-year old olive trees cv. *Manzanillo* during 2013 and 2014 on a loamy sand soil at FiFa farm (km 50 of Cairo-Alexandria road in the north west of Egypt). The trees were cultivated at 8x5 meters distance (105 tree/fed.) and grown under standard cultural practices and subjected to moderate pruning. Drip system irrigation was used (four dippers/tree). Trees were almost uniform in growth and subjected to the same management treatments. Organic manure was applied as compost at rate 40 kg/tree. N, P_2O_5 and K_2O rates of 288, 74 and 192 kg fed^{-1}year, respectively were applied as fertigation. Potassium nitrate (KNO_3) was applied three times in January, February and March as foliar spray. Three foliar sprays of micronutrients were applied during the growing period: before flowering, after fruit set and at the end of summer. Physical and chemical properties of trees rhizosphere soils (Table 2) before treatments application were determined for the two seasons according to Page *et al.* (1982).

TABLE 2. Mechanical and chemical properties of untreated soil.

Character	2013	2014
Particles size distribution		
Crosse sand (%)	4.45	5.41
Fin sand (%)	76.35	78.00
Silt %	12.9	11.08
Clay %	6.30	5.51
Texture	Loamy sand	Loamy sand
Chemical analysis		
EC (dS/m)	4.40	4.30
pH (1:2:5)	8.46	8.10
Organic matter %	0.18	0.26
Organic carbon%	0.11	0.15
Total nutrients ppm		
N	0.13	0.22
P	0.40	0.50
K	1.15	1.62
Mn	1.35	1.90
Zn	0.66	0.80
Fe	1.50	1.82

Experimental field design

The experiments comprised of 7 treatments, 2 trees per each treatment, replicated 3 times in two factorial experiments between application methods and different NPK levels. The trees received the biofertilizer of cyanobacteria and OVW (Cyano/OVW) as soil drench or foliar application.

Soil drench applications

Cyano/OVW biofertilizer was diluted with tap water at the rate of 1:1 (v/v) and was added as soil drench application in the canopy circumference position during the period from first January till the end of fruit set (6 months) each two weeks. Except for control, all treatments received Cyano/OVW as soil drench application at the following rates:

100% = 2 Liters/tree x 6 months x 2 weeks = 24 liter
75% = 3 Liters/tree x 6 months x 2 weeks = 36 liter
50% = 4 Liters/tree x 6 months x 2 weeks = 48 liter

Treatments of soil drench application

- Control of full dose mineral NPK fertilizers and FYM as recommended by the Ministry of Agriculture at the rate of compost 40 kg/tree, N, P_2O_5 and K_2O rates of 288, 74 and 192 kg fed^{-1}year, respectively were applied as fertigation.
- 100% of the recommended mineral NPK fertilizers + 24 liter Cyano/OVW
- 75% of the recommended mineral NPK fertilizers + 36 liter Cyano/OVW
- 50% of the recommended mineral NPK fertilizers + 48 liter Cyano/OVW

Foliar spray application

Farm fertilizer (compost + mineral fertilization) was added at the second week of November and was applied in two parallel ditches of 100x40x30cm, for length, width and depth, respectively surrounded the tree from two directions in the end of canopy shade. 1 liter of Cyano/OVW biofertilizer was diluted with tap water at the rate of 1:4 (5 liter/tree) and was sprayed to each tree of all treatments except for control during the period from first January till the end of fruit set. Cyano/OVW foliar spray rates of 100%, 75% and 50% refer to spraying the four, three and two olive tree directions, respectively.

Treatments of foliar spray application

- Control of full dose mineral NPK fertilizers and FYM as recommended by the Ministry of Agriculture, at the rate of at the rate of compost 40 kg/tree. N, P_2O_5 and K_2O rates of 288, 74 and 192 kg fed^{-1}year, respectively as fertigation.
- 100% of the recommended mineral NPK fertilizers dosage/tree + 100% Cyano/OVW foliar spray (5 liter/tree/all directions).
- 100% of the recommended mineral NPK fertilizers dosage/tree + 75% Cyano/OVW foliar spray (5 liter/tree/three directions).
- 100% of the recommended mineral NPK fertilizers dosage/tree + 50% Cyano/OVW foliar spray (5 liter/tree/two directions, East and West).

Measurements

Soil sampling and analyses

Soil sampling

Soil samples were randomly collected from the zone of the root tips of the trees on the area of tree canopy circumference at the end of each growing season. Depth of the soil sampling was 0-60cm.(Fernandez *et al.*, 1991).

Soil chemical analyses

Air dried soil samples were ground to pass through a 2 mm sieve using a wooden grinding and stored in plastic bottles prior to the physical and chemical analysis. Soil electrical conductivity (EC), soil pH, soluble and total ions were determined according to the methods described by Chapman and Pratt (1961).

Soil enzymes activity

Freshly soil samples of the root zone were tested for dehydrogenase (Casida *et al*., 1964) and nitrogenase (Hardy *et al*., 1973) enzymes activity two weeks after the last biological treatments dose application.

Plant analyses

Vegetative growth

At the end of each growing season (during first week of September) the following characteristics were measured in terms of shoot length (cm), number of leaves per shoot and leaf area (cm^2) using a planimeter according to Aly (2005).

Flowering characteristics

Length of inflorescence

Inflorescence length (cm) was estimated as average inflorescences per tree.

Flowering Density

Twenty shoots per each tree were employed for determine average shoot length and number of inflorescence per one meter was calculated.

Flowering density = No. of inflorescences x100/shoot length (cm)

Number of total flowers/per inflorescence

A sample of twenty inflorescences per tree was used and total number of flowers per inflorescence was counted.

Percentage of perfect flowers

Twenty inflorescences at ballon stage were collected from the middle portions of shoots, from each tree. Number of perfect and total flowers on each inflorescence were recorded and percentage of perfect flowers was calculated (Mofeed, 2002).

The perfect flowers (%) = No. of perfect flowers/No. of total flowersx100

Sex ratio

The ratio of perfect flowers to male flowers was calculated for every replicate (El-Sharony, 2007).

Fruit set and yield

Percentage of fruit set: fruit set percentage at two times first after 21 days from full bloom as initial fruit set and the second 60 days after full bloom as final fruit set according to (Mofeed, 2002).

Fruit set (%) =No. of fruits/No. of total flowers x100

Yield: Average yield per tree was calculated from each treatment as Kg/tree.

Fruits and Stones characterization

Fruits

Fifty fruits per each tree were randomly selected and used to determine the following physical characteristics: Fruit length (cm) - Fruit diameter (cm) - Fruit weight (g)

Stones

Stones were extracted from the selected fruits to determine the following physical characteristics: Stone length (cm), Stone diameter (cm) and Stone weight (g), Flesh weight (g) and flesh/fruit weight (%) were calculated as follows:

Flesh weight= fruit weight- stone weight

Flesh/fruit weight (%) =Flesh weight (g)/Fruit weight x100

Fruit oil content

Fruit oil content (%) as a dry weight was determined according to A.O.A.C. (1995) method by extraction the oil from the dried fruits with soxhelt for extraction apparatus using petroleum ether at 60-80^0 C of boiling point.

Leaf mineral contents

At the first week of August in each season, leaf samples were taken from mid-shoot (Piper, 1950) washed, air dried at 70^{0}C till the constant weight and grounded. The leaf was digested using an acid mixture consisting of nitric, perchloric and sulfuric acids in the ratio of 8:1:1(v/v), respectively for the determination of NPK and microelements as follows:

- Nitrogen was determined by the Microkjeldahl method (Pregl, 1945).
- Phosphorous was estimated by the method of Murphy and Riely (1962).
- Potassium was determined by flame- photometer according to Brown and Lilleland (1946).
- Calcium was determined spectrophotometerically using Atomic Absorption (Perkin Elmer-3300) according to Chapman and Pratt (1961).
- Microelements i.e., Fe, Mn, Zn and Cu (ppm) were spectrophotometerically determined using atomic absorption (Model, spectonic 21D) as described by Jackson (1973).

Economic evaluation

Economic evaluation was calculated according to Heady and Dillon (1961) as follows:

Gross income per treatment (EGP.fed^{-1}) = yield (Kg.fed^{-1}) x price sale of Kg fruit crop

Total cost (EGP.fed^{-1}) = Cost of fertilizers, laborers, irrigation, pesticides and others= 5500 EGP.fed^{-1}

Net return (EGP.fed^{-1}) = Gross income per treatment -total cost

Statistical analysis

The obtained data was subjected to analysis of variance (ANOVA) according to Snedecor and Cochran (1980). Differences between treatments were compared by Duncan's multiple range tests as described in the SAS. (1994).

Results and Discussion

In the present study, the comparative effects of soil drench and foliar spray application methods of Cyano/OVW biofertilizer on soil properties, vegetative parameters, fruiting and yield of olive trees as well as the economic evaluation were investigated.

Cyano/OVW biofertilizer production

The chemical characteristics of the bio-formulated Cyano/OVW showed that the product is almost neutral with low EC and total phenols, very rich in organic matter and macro/micro nutrients (Table 3).

TABLE 3. Chemical characterization of Cyano/OVW biofertilizer.

Characteristics	**Cyano/OVW**
pH	7.50 - 8.50
EC ($dScm^{-1}$)	2.50 – 3.75
Total solids (gl^{-1})	25.00 – 30.00
Chemical Oxygen Demand, COD (gl^{-1})	15.00 – 20.00
Organic matter (%)	2.50 – 3.00
Organic carbon (%)	0.88 – 1.20
Total nitrogen (%)	2.60 – 3.50
Total phosphorus (%)	1.70 – 2.00
Total potassium (%)	2.00 – 3.00
Total Phenols (%)	0.25– 0.30
Soluble nutrients ($mg.l^{-1}$)	
N	1550.0 – 2000.0
P	1000.0 – 1500.0
K	1500.0 – 2000.0
Fe	50.0 – 60.0
Mn	5.0 – 6.0
Zn	2.2– 2.5

Application of biological treatment required diluting the OMW in order to keep the concentration of toxic compounds at a biologically tolerable level for cyanobacteria cultivation (Jaouani *et al.*, 2005). On the other hand, mineral analysis of OMW showed its rich in macro nutrients (NPK) that required for algal growth development. But in our study, dilution maximized the economic value of OMW waste instead of being disposed into the environment without any treatments. It was thus maximized through physical and biological processes that will eventually lead to a new cheap, sustainable and eco-friendly Cyano/OVW biofertilizer.

Effect of Cyano/OVW biofertilizer on soil properties

Enzyme activities and phenolic compounds of soils

Soil enzymes

The enzymatic activities of soils (Table 4) were determined by assessing the activity of dehydrogenase enzyme to provide the correlative information of the biological activity and soil microbial populations and the activity of nitrogenase enzyme as a useful assay for the quantification of the N_2-fixation process in soil (Sherman *et al.*, 2010). In general, application of chemical fertilizers stimulated the growth and multiplication of microorganisms. However, increased dosage inhibits the survival of microbes due to osmotic stress created by fertilizers. Cyano/OVW biofertilizer significantly affected soil enzymes activity compared to control treatment in both seasons. The highest mean values of both enzymes

were recorded by soil drench application. There were no significant differences in dehydrogenase values between 75 and 100% NPK combined with 24 and 36 L of Cyano/OVW, respectively in both seasons. However, 100% NPK and 24 L of Cyano/OVW soil drench application gave the highest nitrogenase activity values in both seasons. It was noticed that foliar spray application had no significant effect on dehydrogenase activity comparing with control during the two seasons. The addition of OVW to soils increased the total microbial population as measured by microbial biomass C and N, soil respiration, and dehydrogenase activity (Mekki *et al.*, 2006 and Di Serio *et al.*, 2008). Whereas, a stimulatory effect of OVW on nitrogen-fixing bacteria was reported by Garcia (2013). On the other hand, Mekki *et al.* (2006) found a significant reduction in the number of soil nitrifying bacteria at the highest OVW dose (400 m^3/ha) applied. Moreover, Di Serio *et al.* (2008) showed that high amounts of OVW increased the soil-denitrifying community and decreased slightly the population of nitrifying bacteria. Therefore, OVW appears to have an effect on different bacterial groups involved in N cycling. The presented results revealed that both soil enzymatic activities (nitrogenase and dehydrogenase enzymes) in these experiments exhibited significant increases due to the enrichment of Cyano/OVW with the selected N_2-fixing cyanobacteria strains (*Anabaena oryzae and Nostoc muscorum*) under different NPK levels. This could be attributed to the great benefits of algal inoculant on soil microbial activity. These findings are supported by our previous investigations (Mostafa *et al.*, 2011, 2013 and 2015) and the results obtained by Rana *et al*. (2012).

Total phenolic compounds in soil

Data in Table 4 revealed that all Cyano/OVW treatments in both application methods reduced total phenols lower than control at both seasons. The highest total phenols mean value of 69.66 mg.Kg^{-1} dry soil was achieved by Cyano/OVW drench application in the first season while, foliar spray application gave the highest mean value of 65.63 mg.Kg^{-1} in the second season. Cyano/OVW foliar spray application recorded the lowest phenols contents of 54.24 and 55.82 mg.Kg^{-1} with 50% Cyano/OVW in the first and second season, respectively. Data showed that the phenols content decreased during the two seasons due to the microflora activity. The untreated OVW application increased the total phenolic compounds content in all soil layers (Mekki *et al.*, 2007). In this study, the reduction of total phenols in soil treated with Cyano/OVW could be due to the highly resistance and biodegradation ability of cyanobacteria to phenolic compounds (EI-Sheekh *et al.*, 2012 and Amores-Sanchez *et al.*, 2015).

Therefore, results emphasized the potential using of cyanobacteria as natural nonpolluting and inexpensive nitrogen and plant growth promoting regulators (BGPR) resource can recycle the nutrients, organic matter and phenols of OVW effluent to sustainable biofertilizer for organic agriculture (Sahraoui *et al.*, 2015).

TABLE 4. Soil enzymes activity and phenolic compounds in soil as affected by Cyano/OVW application methods and NPK levels during 2013 and 2014 seasons.

Treatments		Dehydrogenase (µgTPF.g^{-1} dry soil)		Nitrogenase (µ mol C_2H_4.g^{-1}.h^{-1})		Total phenols mg.Kg^{-1} dry soil	
		2013	2014	2013	2014	2013	2014
Soil Drench application	Control	0.666c	0.697c	13.67g	14.60g	82.69a	76.62a
	100%	3.442a	3.665a	488.01a	521.26a	60.08cd	57.02de
	75%	3.166a	3.414a	178.97b	161.16b	61.66c	58.73cd
	50%	1.860b	1.872b	65.30e	69.75e	74.21b	59.08c
Mean		2.284A	2.412A	186.49A	199.19A	69.66A	62.86B
Foliar spray application	Control	0.666c	0.697c	13.67g	14.60g	82.69a	76.62a
	100%	1.170bc	1.305bc	105.85c	113.07c	58.65d	57.89cd
	75%	0.926c	0.993c	91.45d	97.68d	75.38b	72.17b
	50%	0.677c	0.798c	52.28f	55.84f	54.24e	55.82e
Mean		0.860B	0.948B	65.81B	70.29B	67.74B	65.63A
Mean	Control	0.666C	0.697C	13.67D	14.60D	82.69A	76.62A
	100%	2.306A	2.485A	296.93A	317.16A	59.37D	57.45C
	75%	2.046A	2.204A	135.21B	144.42B	68.52B	65.45B
	50%	1.269B	1.335B	58.79C	62.79C	64.22C	57.45C

Values within same column having different letters showed statistically significant differences ($p<0.005$).

Soil chemical properties

Soil electrical conductivity (EC)

A significant reduction in soil EC was observed in response to Cyano/OVW soil drench and foliar spray methods compared to the control in both seasons (Tables 5 and 6). The raise of soil EC with increasing the untreated OVW rates and the highest OVW dose applied almost duplicates the control salinity. This increase in soil EC was related to the high salts concentration in the OVW. Furthermore, soil sodium adsorption ratio (SAR) and exchangeable sodium percentage (ESP) values were substantially affected by OVW salinity (Chaari *et al.*, 2015). It was recognized that soil drench application reduced soil EC lower than foliar spray in the two seasons. Cyano/OVW caused a significant decrease in soil EC that was due to the presence of cyanobacteria mixed culture. In this respect Al-Sherif *et al.* (2015) reported that soil EC were highly significantly reduced by inoculation with mixed culture of cyanobacteria suspension, *Nostoc minutum* and *Anabaena spiroides*. Fernandes *et al.* (1993) explained that the effect of cyanobacteria in decreasing soil salinity could be due to cyanobacteria exudates which remove Na^+ from aqueous medium by bio sorption thus the osmotic as well as ionic effect of Na^+, which otherwise have an inhibitory effect on growth get a big drop since after betting bound to the cyanobacterial secretion, Na ions are no more available as free ions in the medium. Mahmoud *et al.* (2007) reported that soil inoculation with cyanobacteria led to a significant reduction in sandy soil electric conductivity (EC).

pH of soil

Soil pH in both seasons indicated no significant difference with either application methods or Cyano/OVW doses (Table 5 and 6). Chaari *et al.* (2015) illustrated that regular application of three doses: 50, 100 and 200 $m^3 \cdot ha^{-1}$ of OVW for nine successive years increased the soil electrical conductivity significantly with the increase of OVW rates and pH variations were not detected after ten months of the spreading date. They added that soil pH remains unchangeable mainly due to the presence of organic acids. Chartzoulakis *et al.* (2010) noticed no significant difference in soil pH after 3 years of successive OVW spreading. This result could be explained by the buffering capacity of the soil which counterbalances the negative effect of OVW. Contrary to the work of Di Bene *et al.* (2013), the soil pH variation was noticed between control and treated soil after 6 months of amendment. Whereas, Piotrowska *et al.* (2011) also detected that small pH variations after 2 weeks of OVW treated soil. Mekki *et al.* (2009) demonstrated that the addition of treated or untreated OMW without or after C/N ratio correction did not show any significant effect on the initial soil pH. The also reported that, in spite of the initial untreated OMW acidity, the follow-up of this parameter during 6 months showed that these OMW provoked no significant reduction in the soil pH, whereas the addition of treated OMW provoked a weak augmentation. Similarly, OMW application increased soil EC, and this increase was proportional to the added OMW quantity.

Marco and micronutrients content in soil

Soil contents of N and K revealed that both Cyano/OVW application methods significantly increased total and available records of N, K in both seasons (Tables 5 and 6). The superior concentrations mean values of the available N and K in soil were detected with treatments of soil drench application in both seasons. The highest values of these elements were recorded with tress received 100% NPK combined with 24 L/tree (100%) of Cyano/OVW in both seasons. Regarding P concentrations in soil, the available concentrations of P in soil were significantly enhanced by both Cyano/OVW application methods at the first season (Table, 5). 100% Cyano/OVW (24L/tree) soil drench application and 100% NPK recorded the highest significant available P value (9.80 $mg.kg^{-1}$). However, available P indicated no significant difference with either application methods or Cyano/OVW rates at the second season (Table 6). As for total phosphorus in soil as affected by Cyano/OVW biofertilizer, both of Cyano/OVW application methods and rates with any of NPK levels had no significant effects on total phosphorus (%) in soil. In this respect, Sahu *et al.* (2012) reported that cyanobacteria biofertilizers convert insoluble phosphorus in soil into forms available to plants, thereby increasing their efficiency and availability. These microalgae caused higher P content due to their ability to produce organic acid such as gluconic, citric, and fumaric acids under P-limiting conditions. In this study, the insignificant decrease in soil pH values due to Cyano/OVW biofertilizer may be increased the availability of P in soil. These results are in line with those of Grzesik and Romanowska-Duda (2015) who found that the phosphatases are considered also to be a good indicator of organic phosphorus mineralization potential and biological activity of soil. The availability of

micronutrients in soil revealed different trends depending on Cyano/OVW biofertilizer application methods and rates at different NPK levels during the two studied seasons (Tables 5 and 6). At the first season, the highest available means of Mn (1.64 ppm) and Zn (0.91 ppm) achieved by Cyano/OVW soil drench application. Best results of Mn (1.90 ppm) and Zn (1.0 ppm) were obtained with 100% NPK combined with 100% Cyano/OVW soil drench application. While, application methods of Cyano/OVW biofertilizer had no significant effect on the available Fe in soil. However, NPK at 75% recorded the highest mean available Fe value (3.10 ppm).

TABLE 5. Chemical properties of soil as affected by Cyano/OVW biofertilizers during season of 2013.

Treatments		EC ds/m	pH	Available (ppm)			Total (%)			Available (ppm)			OM %	OC %	C/N ratio
				N	P	K	N	P	K	Mn	Zn	Fe			
Soil Drench application	Control	4.46a	8.46a	33.40de	8.65b	237.4e	0.16de	0.037a	0.104d	1.40c	0.78b	2.22d	0.502a	0.292c	1.79b
	100%	2.54d	8.00a	53.01a	9.80a	522.5a	0.26a	0.039a	0.23a	1.90a	1.00a	2.37d	0.610a	0.355a	1.38d
	75%	3.47c	8.16a	34.77cd	9.45ab	482.2b	0.17c-e	0.039a	0.20b	1.57bc	0.93ab	3.56a	0.600a	0.349a	2.04a
	50%	3.96b	8.04a	35.74bc	9.11ab	388.4d	0.19b	0.037a	0.20b	1.69b	0.91ab	2.27d	0.571a	0.332b	1.73bc
Mean		3.61A	8.17a	39.23A	9.25A	407.6A	0.20A	0.038A	0.18A	1.64A	0.91A	2.61A	0.571A	0.332A	1.74B
Foliar spray application	Control	4.46a	8.46a	33.40de	8.65b	237.4e	0.16de	0.037a	0.104d	1.40c	0.78b	2.22d	0.502a	0.292c	1.79b
	100%	0.86e	8.18a	37.56 b	9.00ab	480.8b	0.19bc	0.038a	0.19c	1.58bc	0.90ab	2.86b	0.531a	0.309c	1.65c
	75%	0.92e	8.02a	31.65e	9.25ab	473.6c	0.16e	0.037a	0.20b	1.19d	0.62c	2.64bc	0.561a	0.326b	2.08a
	50%	1.00e	8.01a	31.65e	7.15c	391.9d	0.18b-d	0.037a	0.20b	0.81e	0.47d	2.43cd	0.522a	0.303c	1.71bc
Mean		1.81B	8.17a	33.57B	8.51B	395.9B	0.17B	0.037A	0.17B	1.25B	0.69B	2.54A	0.529A	0.308B	1.81A
Mean	Control	4.46A	8.46A	33.40B	8.65AB	237.4D	0.16C	0.037A	0.10B	1.40B	0.78B	2.22C	0.502A	0.292C	1.79B
	100%	1.70C	8.09A	45.28A	9.40A	501.7A	0.22A	0.038A	0.21A	1.74A	0.95A	2.62B	0.571A	0.332A	1.52C
	75%	2.19B	8.09A	33.21B	9.35A	477.9B	0.16C	0.038A	0.20A	1.38BC	0.78B	3.10A	0.581A	0.338A	2.06A
	50%	2.48B	8.03A	33.69B	8.13B	390.2C	0.19B	0.037A	0.20A	1.25C	0.69B	2.35C	0.546A	0.318B	1.72B

Values within same column having different letters showed statistically significant differences ($p<0.005$)

The best integration was found by treatment that combining Cyano/OVW soil application method (36L/tree) and NPK 75% (3.65 ppm). Regarding the second season, Mn gave the same trend for the first season. On the other hand, neither application methods/rates nor NPK% significantly affected available Zn concentration in the second season. Foliar spray application method of Cyano/OVW significantly increased the mean value of available Fe (2.77 ppm) particularly with 75% NPK and 36 L/tree foliar spray application. Boraste *et al.* (2009) also reported that biofertilizer enhance the nutrient availability to crop plants (by process like fixing atmosphere N or dissolving P present in the soil), and impart better health to plants and soil thereby enhancing crop yields in

moderate way. Our study indicated that Cyano/OVW biofertilizer, being essential components of organic farming, could play a vital role in maintaining long-term soil fertility and sustainability by fixing atmospheric dinitrogen (N=N), mobilizing fixed macro and micro nutrients (Sahu *et al.*, 2012).

TABLE 6. Chemical properties of soil as affected by Cyano/OVW biofertilizer during season of 2014.

Treatments		EC ds/m	pH	Available (ppm)			Total (%)			Available (ppm)			OM %	OC %	C/N ratio
				N	P	K	N	P	K	Mn	Zn	Fe			
Soil Drench application	Control	4.39a	7.96a	36.54ef	7.96a	91.35f	0.127e	0.036a	0.038c	1.40c	0.78b	2.22d	0.580b	0.337b	2.658a
	100%	2.61c	8.05a	52.21a	8.11a	149.25a	0.288a	0.041a	0.074a	1.90a	1.00a	2.37d	0.620a	0.360a	1.251d
	75%	2.02d	7.91a	43.56c	7.91a	143.07b	0.212c	0.034a	0.063ab	1.57bc	0.93ab	3.56a	0.610a	0.355a	1.670bc
	50%	3.41b	8.00a	46.11b	8.00a	141.54b	0.247b	0.029a	0.071a	1.69b	0.91ab	2.27d	0.610a	0.355a	1.435cd
Mean		3.11A	7.98A	44.60A	7.99A	131.30A	0.219A	0.035A	0.061A	1.64A	0.91A	2.61A	0.605A	0.352A	1.754B
Foliar spray application	Control	4.39a	7.96a	36.54ef	7.96a	91.35f	0.127e	0.036a	0.038c	1.40c	0.78b	2.22d	0.580b	0.337b	2.658a
	100%	0.28e	7.88a	38.18de	7.88a	105.00d	0.186d	0.034a	0.046bc	1.58bc	0.90ab	2.86b	0.540c	0.314c	1.687bc
	75%	0.77e	8.00a	35.80f	8.00a	96.04e	0.175d	0.034a	0.041c	1.19d	0.62c	2.64bc	0.570b	0.331b	1.897b
	50%	0.75e	8.00a	39.19d	8.00a	115.86c	0.192d	0.033a	0.050bc	0.81e	0.47d	2.43cd	0.510d	0.297d	1.569c
Mean		1.55B	7.96A	37.38B	7.96A	102.06B	0.17B	0.037A	0.17B	1.25B	0.69B	2.54A	0.550B	0.320B	1.953A
Mean	Control	4.39A	7.96A	36.54D	7.96A	91.35C	0.16C	0.037A	0.10B	1.40B	0.78B	2.22C	0.580A	037AB	2.658A
	100%	1.45C	7.99A	45.19A	7.99A	127.13A	0.22A	0.038A	0.21A	1.74A	0.95A	2.62B	0.580A	0.337AB	1.469C
	75%	1.40C	7.99A	39.68C	7.99A	119.55B	0.16C	0.038A	0.20A	1.38BC	0.78B	3.10A	0.590A	0.343A	1.784B
	50%	2.08B	8.00A	42.65B	8.00A	128.70A	0.19B	0.037A	0.20A	1.25C	0.69B	2.35C	0.560B	0.326B	1.502C

Values within same column having different letters showed statistically significant differences ($p<0.005$)

Organic matter (OM), organic carbon (OC) and C/N ratio of soil

Concerning organic matter (OM) content in soil at the first season, no significant effect was found on OM values in response to Cyano/OVW application methods and doses (Table 5). While, Cyano/OVW soil drench application and 50, 75 and 100% NPK gave the similar highest significant records of organic matter at the second season (Table 6). The highest C/N ratio of 2.04 and 2.08 were observed with soil drench application at the rate of 36 L/tree of Cyano/OVW + 75% NPK and with 75% foliar spray respectively at the first season (Table 5). While, in the second season the highest values were recorded by control treatment (Table 6). Mekki *et al.* (2009) reported that the increase of soil organic matter rate was proportional to the increasing of OVW doses application. The soil organic matter increased from 0.068% in the control to 0.2%, 0.35% and 0.5% with OVW gradual doses application 50, 100 and 200 $m^3 \cdot ha^{-1}$, respectively. Also, a slight increase in soil organic matter was observed due to inoculation with cyanobacteria. These results are in harmony with those of Acea *et al.* (2003) who showed that inoculation with different

cyanobacteria strains induced great microbial proliferation as well as high increases in soil organic matter that leads also to increase the soil microorganisms' population. The micronutrients content in soil increased with increasing soil OM content, thus showing the importance of soil OM in micronutrient availability for crops (Lia *et al.*, 2007). Also, Palaniappan *et al.* (2010) explained that cyanobacterial extract is known to improve the soil stability due it containing polysaccharides and lipids which add organic matter to the soil and hence, aid in enhancing soil aggregation that improved the nutrients availability preventing them from loss from soil. Nitrogen fixers break down enzymes containing nitrogen that are not readily accessible to plants. As a result of this breakdown, nitrogen, the most important element to plant growth, becomes readily available to take up by plant. Upon decomposition, blue green algae provide fresh organic matter which in turn helps to promote the growth of beneficial microorganisms that further help make nutrients readily available to plants (Yoshida and Ancajas, 1973). Results in this text revealed that several chemical and biochemical properties of the investigated soils changed in response to Cyano/OVW soil drench and foliar spray applications and rates. In this respect, the presented data drawing the attention to asses different treatments methods, as previously mentioned, have to be proposed to solve the problems associated with OVW in this study.

Olive vegetative growth parameters

It was obvious that shoot growth parameters (shoot length and number of leaves/shoot) were significantly influenced by different application methods and NPK rates compared to the control during both seasons (Table 7). Leaf area and shoot growth parameters (shoot length and number of leaves/shoot) of *Manzanillo* olive cultivar were significantly affected by Cyano/OVW biofertilizer application methods. The highest leaf area at the first season (3.43 cm^2) was recorded by soil drench application method combined with 100% NPK while, foliar spray application at 50% NPK achieved the highest value (4.06 cm^2) at the second season.

However, the highest shoot length and number of leaves/shoot were obtained by foliar spray application with 50% NPK in both seasons. Hence the highest shoot length was obtained for trees received the rate of 100% OVW soil drench application (23.04 and 25.50 cm) in both seasons, respectively. Concerning foliar spray application, the height shoot length was significantly for trees received the rate of 50% (28.73 and 33.30cm) in both seasons. The highest mean values of shoot length (24.27 and 29.18 cm) were obtained for trees received 50% OVW foliar spray in both seasons, respectively. Osman *et al.* (2010) revealed that bio and NPK fertilizer treatments significantly increase number of shoots/ branch/ meter, number of leaves per shoot, shoot length, shoot diameter, leaf area, leaf fresh and dry weights, N,P and K contents in olive leaves. Also, Al-Absi (2010) suggested that the high concentration of olive waste water used, significantly increased the young shoot length, number of new shoot and leaf area of Nabali and *Manzanillo* trees than of the control. Foliar application of algae extract was accompanied with stimulating growth characters significantly compared to untreated vines. The promotion

was depended on increasing algal extract concentrations. The increase in such characters was insignificant when algal extract was used at concentrations higher than 50% (Abd El-Monium and Abd-allah, 2008). Cyanobacteria contain growth regulators (auxins, cytokinins and gibberellins), betain, amino acids and inorganic elements that influence cell growth and division cycle, expansion, nutrition and maturity (Nanjappan *et al.*, 2007).

TABLE 7. Effect of Cyano/OVW application methods and NPK levels on leaf area, shoot length and number of Manzanillo olive leaves during 2013 and 2014 seasons.

Treatments		Leaf area (cm^2)		Shoot length(cm)		No. of leaves	
		2013	2014	2013	2014	2013	2014
Soil drench application	Control	2.34d	2.58e	17.20e	19.73g	15.65f	16.00g
	100%	3.43a	3.52c	23.04b	25.50c	23.76b	24.60c
	75%	3.12b	3.26d	17.77e	21.90f	19.29e	19.60f
	50%	3.14b	3.47c	19.80c	25.07d	21.60c	21.80d
Mean		3.01A	3.21B	19.45B	23.05B	20.08B	20.50B
Foliar spray application	Control	2.34d	2.58e	17.20e	19.73g	15.65f	16.00g
	100%	2.58c	2.62e	18.90d	22.80e	19.75d	20.70e
	75%	3.15b	3.94b	20.27c	28.50b	24.52a	25.40b
	50%	3.15b	4.06a	28.73a	33.30a	24.54a	25.90a
Mean		2.80B	3.30A	21.27A	26.08A	21.12A	22.00A
Mean	Control	2.34C	2.58D	17.20D	19.73D	15.65D	16.00D
	100%	3.00B	3.07C	20.97B	24.15C	21.76C	22.65B
	75%	3.14A	3.60B	19.02C	25.20B	21.91B	22.50C
	50%	3.14A	3.77A	24.27A	29.18A	23.07A	23.85A

Values within same column having different letters showed statistically significant differences ($p<0.005$)

Leaf mineral content

The effect of Cyano/OVW applications and different NPK levels on macro (N, P and K%) and micro (Mn, Zn and Fe ppm) elements content in *Manzanillo* olive leaves was shown in Tables (8 and 9).

Nitrogen

Nitrogen content in olive leaves was significantly affected by treatments in both seasons. However, both soil drench and foliar spray application methods of Cyano/OVW tended to increase N % in the leaves compared to the control. The highest N percentages in leaves (2.56 and 2.88%) were obtained by Cyano/OVW soil drench application with 100% NPK in both seasons, respectively.

Phosphorus

Data presented in Tables (8 and 9) show that Cyano/OVW biofertilizer application methods and doses had no significant effect on phosphorus percentage in the leaves of *Manzanillo* olive trees differ during the two seasons.

Potassium

Soil drench application of Cyano/OVW significantly increased K content in leaves higher than foliar spray application in both seasons. This may be due to encourage of K absorption from soil rather than utilization in plant tissues (Abd El-Motty *et al.*, 2010). K was significantly affected by the OVW treatment at both spreading sites and just after OVW disposal with values was found to be four to 10 folds higher than the observed ones in the control (Di Bene *et al.*, 2013). Magdich *et al.* (2013) has reported a significant increase of soil K concentration with a value of around 985 ppm at the surface layer (0–20 cm) of the soil treated with dose 200 m^3/ha/ yr of OVW. Accordingly, this level was almost 6 times higher than that in the control. These results provide support for the proposals in the literature the candidacy of OVW for application as an alternative K fertilizer (Di Serio *et al.*, 2008).

Micronutrients and phenols contents of leaves

Concerning micronutrients (Mn, Zn and Fe) in leaves, results cleared that all studied treatments significantly increased the micronutrients content of leaves in both seasons (Tables 8 and 9). 100% Cyano/OVW soil drench application was the superior treatment and recorded the highest Mn (19.00 and 22.00 ppm) and Zn (10.0 and 11.50 ppm) values in seasons of 2013 and 2014, respectively. As for Fe, 75% Cyano/OVW soil drench application was the superior treatment of 35.60 ppm Fe content of leaves in the first season while, Cyano/OVW foliar spray at the rate of 75% recorded the highest Fe content (29.40 ppm) of leaves in the second season. The positive effects of Cyano/OVW could be attributed to the effect of cyanobacteria to improve the availability of metals and increase the levels of extractable N, P, K, Fe, Zn and Mn than control for uptake by plants and also improve the physical characteristics of the soil, which greatly influenced growth and absorption of many elements needed to the plant growth (El-Karamany *et al.*, 2000). This may help minimizing the amounts of chemical fertilizers and improve their application efficiency and subsequently avoiding environmental pollution by the access of these chemicals. In addition to their role in enhancing the growth of the plants, bio-fertilizers can act as biocontrol agents in the rhizosphere at the same time. Abd El Monium and Abd-allah (2008) reported that a progressive increase of percentages of N, P, and K in leaves was observed as a result of increasing concentration of algal extract till 50% on Superior grapevines.

Flowering Characteristics

Length of inflorescences

Data in Table 10 revealed that all treatments gave the significantly higher length of inflorescences than control which recorded the least significant values during the two seasons. Trees treated with 50% Cyano/OVW foliar spray application gave the highest length of inflorescences (3.06 and 3.69 cm) in both studied seasons.

TABLE 8. Effect of Cyano/OVW application methods and NPK levels on some macro, micro nutrients and phenols of Manzanillo olive leaves during 2013 season.

Treatments		(%)			(ppm)			
		N	P	K	Mn	Zn	Fe	Phenols
Soil Drench application	Control	1.63de	0.37a	1.04d	14.00c	7.80b	22.20d	25.50a
	100%	2.56a	0.39a	2.28a	19.00a	10.00a	23.70d	20.95c
	75%	1.71c-e	0.39a	1.99b	15.70bc	9.30ab	35.60a	23.49b
	50%	1.92b	0.37a	1.95b	16.90b	9.10ab	22.70d	14.47d
Mean		1.96A	0.38A	1.82A	16.40A	9.10A	26.10A	21.10 B
Foliar spray application	Control	1.63de	0.37a	1.04d	14.00c	7.80b	22.20d	25.50a
	100%	1.88bc	0.38a	1.88c	15.80bc	9.00ab	28.60b	20.74c
	75%	1.57e	0.37a	1.96b	11.90d	6.20c	26.40bc	21.41c
	50%	1.78b-d	0.37a	1.95b	8.10e	4.70d	24.30cd	22.16c
Mean		1.71B	0.37A	1.71B	12.50B	6.90B	25.40A	22.45 A
Mean	Control	1.63C	0.37A	1.04B	14.00B	7.80B	22.20C	25.50A
	100%	2.22A	0.38A	2.08A	17.40A	9.50A	26.20B	20.85C
	75%	1.64C	0.38A	1.98A	13.80BC	7.80B	31.00A	22.45B
	50%	1.85B	0.37A	1.95A	12.50C	6.90B	23.50C	18.32D

TABLE 9. Effect of Cyano/OVW application methods and NPK levels on some macro, micro nutrients and phenols of Manzanillo olive leaves during 2014 season.

Treatments		(%)			(ppm)			
		N	P	K	Mn	Zn	Fe	Phenols
Soil Drench application	Control	1.30e	0.40a	0.38c	19.00b-d	9.90c	27. 40ab	28.00a
	100%	2.88a	0.41a	0.74a	22.00a	11.50a	28.80ab	17.03f
	75%	2.12c	0.34a	0.63ab	19.40b-d	9.10d	25.60b	25.50b
	50%	2.47b	0.29a	0.71a	19.90bc	10.40bc	21.80c	18.05ef
Mean		2.19A	0.35A	0.61A	20.10A	10.20A	25.90B	22.15 A
Foliar spray application	Control	1.27e	0.36a	0.38c	19.00b-d	9.90c	27.40ab	28.00a
	100%	1.86d	0.34a	0.46bc	18.40cd	8.80d	27. 40ab	24.16c
	75%	1.75d	0.34a	0.41c	20.50ab	10.80ab	29. 40a	18.23e
	50%	1.92d	0.33a	0.50bc	17.80d	10.00c	26.50ab	20.49d
Mean		1.70B	0.34A	0.43B	18.90B	9.90A	27.70A	22.72 A
Mean	Control	1.27D	0.36A	0.38B	19.00AB	9.90A	27.40A	28.00A
	100%	2.37A	0.37A	0.60A	20.20A	10. 20A	28.10A	20.59C
	75%	1.94C	0.34A	0.52A	20.00AB	10.00A	27.50A	21.86B
	50%	2.18B	0.31A	0.60A	18.90B	10.20A	24.20B	19.27D

Flowering density

Flowering density as measured by number of inflorescences per meter responded significantly to all treatments used in both studied seasons as compared with control trees which exhibited the least average number of inflorescences (Table 10). Accordingly, flowering density had the same trend of length of inflorescences as the highest flowering density of 24.70 and 35.80 were recorded by 50% Cyano/OVW foliar spray application in the first and second seasons, respectively while, the lowest records of 15.43 and 15.70 were observed with control treatments during the first and second seasons, respectively.

Number of total flowers/inflorescences

Table 10 showed that in general Cyano/OVW biofertilizer application significantly increased the number of total flowers/inflorescences in both seasons over than control. Whereas the highest numbers of total flowers/inflorescences of 14.33 and 16.00 were obtained by 50% Cyano/OVW foliar spray application in both seasons, respectively.

Perfect flowers

Percentage of perfect flowers was significantly increased with the application of Cyano/OVW biofertilizer while, the untreated trees gave the lowest perfect flowers % of 43.80 and 45.80 in both seasons, respectively. As for spray application, the trees treated with 50% Cyano/OVW biofertilizer gave the highest perfect flowers percentages of 63.07 and 75.30 in both seasons, respectively

Sex ratio

As for sex ratio, percentage of sex expression ratio of *Manzanillo* olive trees as affected by treatments was illustrated in Table 10. It was observed that the trees treated with 50% Cyano/OVW biofertilizer foliar spray application gained the highest sex ratio of 3.00 and 3.57 in both seasons, respectively. Referring to the previously results one can see that all flowering measurement were significantly higher with Cyano/OVW biofertilizer as compared with control. This improvement in flowering measures resulted may be attributed to the stimulation effect of the absorbed nutrients on photosynthesis process, particularly with foliar spray application, which certainly reflected positively on both vegetative growth and flowering characteristics (Hegazi *et al.,* 2007).

TABLE 10. Effect of Cyano/OVW on flowering characteristics of Manzanillo olive trees during 2013 and 2014 seasons.

Treatments		Length of inflorescences (cm)		Flowering density		No. of total flowers/infl.		Perfect flowers		Sex ratio	
		2013	2014	2013	2014	2013	2014	2013	2014	2013	2014
Soil drench application	Control	1.80f	2.08g	15.43g	15.70g	9.30f	10.74g	43.80g	45.80g	0.65f	0.47f
	100%	2.44c	2.71c	25.50b	26.27c	11.80b	13.50c	58.50b	71.30c	2.64b	2.63b
	75%	1.90e	2.20f	18.87f	19.67f	9.57ef	11.67f	48.73f	49.79f	1.61d	0.64e
	50%	2.20d	2.50d	20.70d	25.10d	10.20d	13.23d	56.80d	66.07d	1.92c	1.97c
Mean		2.09B	2.37B	20.12B	21.68B	10.22B	12.28B	51.96B	58.24B	1.71A	1.43 B
Foliar spray application	Control	1.80f	2.08g	15.43g	15.70g	9.30f	10.74g	43.80g	45.80g	0.65f	0.47f
	100%	1.96e	2.35e	19.53e	22.40e	9.67e	12.40e	55.40e	56.71e	0.83e	1.07d
	75%	2.71b	3.10b	24.33c	32.40b	10.73c	14.70b	56.97c	74.30b	2.06c	2.56b
	50%	3.06a	3.69a	24.70a	35.80a	14.33a	16.00a	63.07a	75.30a	3.00a	3.57a
Mean		2.38A	2.80A	23.50A	26.58A	11.01A	13.46A	54.81A	63.03A	1.63A	1.92A
Mean	Control	1.80D	2.08D	15.43D	15.70D	9.30D	10.74D	43.80D	45.80A	0.65C	0.47D
	100%	2.20C	2.53C	22.52B	24.33C	10.73B	12.95C	56.95B	64.01B	1.74B	1.85B
	75%	2.31B	2.65B	21.60C	26.63B	10.15C	13.18B	52.85C	62.04C	1.83B	1.60C
	50%	2.63A	3.10A	27.70A	30.45A	12.27A	14.62A	59.93A	70.68A	2.40A	2.77A

Values within same column having different letters showed statistically significant differences ($p<0.005$)

Maksoud (2000) reported that all organic manures applied to olive trees produced increases in number of inflorescences/shoot and number of flowers/ inflorescence. In addition, these results are nearly in the same line with those obtained by Abou Taleb *et al.* (2004) who indicated that inoculation with *Bacillus* +500g N were the most effective in enhancing total number of flowers /inflorescent and sex ratio of olive trees. Also Hegazi *et al.* (2007) found that use different organic fertilization such as poultry manure gave a significant increase of flowering density and sex ratio of olive tree CV Picual.

Fruiting and yield

Initial fruit and final fruit set

Data in Table 11 revealed that all the tested treatments significantly increased initial fruit and final fruit set percentages. The highest initial (6.80 and 8.15%) and final (2.91 and 5.81%) fruit set were achieved by foliar spray application of Cyano/OVW biofertilizer at the rate of 50% during 2013 and 2014 seasons, respectively. On other hand, the untreated trees had the lowest initial and final fruit set in both seasons. The previous results are in harmony with that obtained by Hegazi and Stino (1982) on olive Picual cv. who reported that the bud burst, flower bud formation and percentage of perfect flowers was significantly increased by kinetin 100 mg/L. It can be suggested that the high concentrations of certain cytokinin levels may have a positive effect on flower formation in olive during the induction and initiation periods (Ulger *et al.*, 2004). Moreover, probably benzyladenine as a cytokinin compound delayed the senescence stages of buds and increased the entrance of photosynthetic compounds, hormones and other metabolites to inflorescence buds, which are so important for preventing bud abscission and increased the fruit set (Alireza *et al.*, 2006).

Fruit weight and Yield

During the first season, the highest significant fruit weight of 6.45 g resulted from soil drench application of Cyano/OVW biofertilizer at the rate of 50% while, the highest record of 4.93 g in the second season was obtained by foliar spray application of Cyano/OVW biofertilizer at the rate of 75%. Meanwhile, the lowest significant fruit weight was produced from trees treated with Cyano/OVW biofertilizer foliar spray application at the rates of 50% and 100% as it reached only 4.96 and 4.05 g in the first season and second seasons, respectively lower than control treatment (Table 11).

Data in Table 11 illustrated that all Cyano/OVW treatments had a pronounced increase of *Manzanillo* trees yield (kg/tree). Foliar spray application at the rate of 50% recoded the highest significant increase in yield reached 24.53 and 40.37 Kg/tree during the first and second seasons, respectively. Compared to the lowest significant yield produced from the control trees (8.74 and 17.47 Kg/tree) in the first and second seasons, respectively. Algae have a positive effect on fruit setting, yield and fruit quality (Hegab *et al*., 2005). Cyanobacteria extract as a new bio fertilizer containing N, P, K, Ca, Mg, and S as well as Zn, Fe, Mn, Cu, Mo, and Co, some growth regulators, polyamines and vitamins applied to improve nutritional status, vegetative growth, yield and fruit quality in different

orchard as well as vineyards (Abd El-Moniem and Abd-Allah, 2008). Kulk (1995) and Adam (1999) reported the growth promotion in response to application of nitrogen fixer cyanobacterium (*Nostoc muscorn*) could be attributed to the nitrogenase as well as nitrate reductase activities of algae associated with the surface of plants, or the amino acids and peptides produced in algal filtrate and / or other compounds that stimulated growth of crop plants.

TABLE 11. Effect of Cyano/OVW biofertilizer on fruit set and yield of Manzanillo olive trees during 2013 and 2014 seasons.

Treatments		Initial fruit set %		Final fruit set %		F. weight (g)		Yield (Kg/tree)	
		2013	2014	2013	2014	2013	2014	2013	2014
Soil drench application	Control	4.28e	5.06e	1.87e	2.18d	6.00d	4.35f	8.74g	17.47g
	100%	4.93c	7.33b	2.54b	4.35b	5.45e	4.61e	22.63b	32.10c
	75%	4.29e	5.60d	1.96de	2.77c	6.20c	4.64d	9.50f	20.67f
	50%	4.83cd	7.22b	2.18cd	4.22b	6.45a	4.75c	11.33d	30.00d
Mean		4.58B	6.30B	3.14B	3.38B	6.03A	4.59A	13.05B	25.06B
Foliar spray application	Control	4.28e	5.06e	1.87e	2.18d	6.00d	4.35f	8.74g	17.47g
	100%	4.51de	6.73c	2.02de	4.14b	5.16f	4.05g	10.07e	28.47e
	75%	5.74b	7.84a	2.43bc	5.70a	6.30b	4.93a	16.67c	35.50b
	50%	6.80a	8.15a	2.91a	5.81a	4.96g	4.80b	24.53a	40.37a
Mean		5.33A	6.94A	2.31A	4.46A	5.61B	4.53B	15.00A	30.45A
Mean	Control	4.28D	5.06D	1.87C	2.18C	6.00B	4.35C	8.74D	17.47D
	100%	4.72C	7.03B	2.28B	4.24B	5.30D	4.33D	16.35B	30.28B
	75%	5.02B	6.72C	1.19B	4.24B	6.25A	4.79A	13.08C	28.08C
	50%	5.81A	7.69A	2.55A	5.02A	5.71C	4.77B	17.93A	35.18A

Values within same column having different letters showed statistically significant differences ($p<0.005$)

Fruit quality

The effect of the tested treatments on fruit length, diameter, shape index, flesh weight and flesh/ fruits of *Manzanillo* olive trees are shown in Table 12. In this respect, the highest mean value of fruit length (2.65 cm) was recorded by control treatment in the first season while, 75% NPK fertilizer with 36 liter/tree of Cyano/OVW gave the highest mean value (2.33 cm) in the second season. As for Cyano/OVW application methods, foliar spray application method recorded the highest mean value of fruit length (2.51 cm) and its highest record was found in the control treatment (2.65 cm) at the first season. On contrary, in the second season soil drench application was superior in fruit length mean value (2.26 cm) however, both soil drench application at 100% and foliar spray at 75% gave the similar highest value of fruit length (2.35 cm). Fruit diameter of *Manzanillo* olive trees increased significantly as affected by different methods and rates of Cyano/OVW. Data presented in Table 12 showed that soil drench application recorded the highest fruit diameter mean values of 2.27 and 2.08 cm at both seasons, respectively. The most effective treatment was found with trees that received 36 L/tree liter Cyano/OVW and 75% NPK fertilizer in both seasons however, 50% NPK with 48 L/tree Cyano/OVW gave the similar significant highest fruit diameter in the second season. As for application rates the control and Cyano/OVW at rate of 75% gave the similar highest fruit diameter mean value of 2.30 and 2.27 cm, respectively in

the first season, while 50% and 75% NPK with 36 and 48 L/tree recorded the highest mean values of 2.11 and 2.10 cm, respectively in the second one. Two application rates of 50 and 75% NPK combined with 36 and 48 L/tree recorded the similar highest fruit diameter (2.25 and 2.25 cm) in the second season. The present data are in line with many other workers, El-Taweel (2005) who reported that new organic fertilizers can provide plants with an adequate nutrient supply and could be used successfully in organic apple nurseries. The positive influence of rhizosphere microorganisms on nutrient uptake is well established and Abd El-Motty *et al.* (2010) also had estimated the effect of algae extract and yeast application on fruit quality of Keitte mango trees.

Fruit shape index was significantly affected by different treatments in both seasons. Soil drench application of Cyano/OVW at 24 L/tree + 100% NPK fertilizer gave the highest fruit shape index (1.23 and 1.23) in both seasons. Regarding application methods, fruit shape index was significantly increased by foliar application than soil drench application in both seasons. Meanwhile, 100% NPK either by soil drench or foliar spray application recorded the same highest fruit shape index of 1.23 in the first season same highest significant fruit shape index (1.22) was achieved by 50% foliar spray with Cyano/OVW at 48 liter in the first season.

As for flesh fruit weight (Table, 12), the trends showed that soil drench application was the most effective method resulted in a significant increase in flesh weight (5.14 and 3.72 g) in both seasons. The highest flesh fruit weight of 5.36 and 3.94 g were recorded by trees that received 75 and 50% NPK fertilizer with Cyano/OVW at rates of 36 and 48 liter per tree in the both seasons, respectively.

TABLE 12. Effect of Cyano/OVW biofertilizer on fruit quality of Manzanillo olive trees during 2013 and 2014 seasons.

Treatments		Fruit length (cm)		F. diameter (cm)		F. shape index (L/D)		Flesh fruit weight (g)		Flesh/fruits	
		2013	2014	2013	2014	2013	2014	2013	2014	2013	2014
Soil drench application	Control	2.65a	2.20c	2.30b	1.90c	1.15b	1.16bc	5.06d	3.33f	84.29cd	76.62c
	100%	2.55b	2.33a	2.08d	1.90c	1.23a	1.23a	4.64e	3.86d	85.13b	83.66a
	75%	2.41d	2.24b	2.40a	2.25a	1.00c	1.00d	5.38b	3.81e	86.78a	82.13b
	50%	2.31e	2.25b	2.31b	2.25a	1.00c	1.00d	5.48a	3.88c	84.96bc	81.67b
Mean		2.48B	2.26A	2.27A	2.08A	1.10B	1.10B	5.14A	3.72A	85.29A	81.02A
Foliar spray application	Control	2.65a	2.20c	2.30b	1.90c	1.15b	1.16bc	5.06d	3.33f	84.29cd	76.62c
	100%	2.45c	2.20c	2.20e	1.89c	1.23a	1.17bc	4.35f	3.30g	84.36b-d	81.50b
	75%	2.55b	2.35a	2.14c	1.97b	1.19ab	1.20ab	5.33c	4.03a	84.65b-d	81.62b
	50%	2.40d	2.17d	1.97e	1.94bc	1.22a	1.12c	4.17g	4.00b	83.95d	83.35a
Mean		2.51A	2.23B	2.10B	1.92B	1.20A	1.16A	4.73B	3.67B	84.31B	80.77A
Mean	Control	2.65A	2.20C	2.30A	1.90B	1.15B	1.16B	5.06B	3.33D	84.29B	76.62C
	100%	2.50B	2.27B	2.04C	1.90B	1.23A	1.20A	4.49D	3.58C	84.74B	82.58A
	75%	2.48C	2.30A	2.27A	2.11A	1.10C	1.10C	5.36A	3.92B	85.72A	81.88B
	50%	2.36D	2.21C	2.14B	2.10A	1.11C	1.06C	4.82C	3.94A	84.46B	82.51A

Values within same column having different letters showed statistically significant differences ($p<0.005$)

Regarding flesh/ fruit weight percent, soil drench application method was superior to foliar spray at the first season while both application methods had no significant effect on flesh/ fruit weight percent in the second season. Cyano/OVW at rates of 36 and 48 liter/tree combined with 75 and 50% NPK fertilizer resulted in the highest flesh/ fruit of 85.72 and 82.85% in both seasons, respectively. The most effective treatments of 86.78 and 83.35% were due to soil drench application of 36 liter/tree Cyano/OVW + 75% NPK fertilizer in the first season and to foliar spray application of 48 liter/tree Cyano/OVW + 50% NPK fertilizer in the second season, respectively. These results are in harmony with those obtained by Abd El Monium and Abd-allah (2008) and Abd El-Motty *et al.* (2010). Also, Vossen (2007) reported that all the necessary nutrients for olive trees are available via ground application, and the effect is much more lasting. Research trials have proven foliar spray of nitrogen, boron, and potassium will be rapidly taken up by the tree. Nutrient levels will increase dramatically in the leaves, but only for a short while, usually just a few weeks. There is no measurable effect on shot berries, fruit set, shoot growth, number of flowers, number of perfect flowers, fruit yield, or fruit size. The best plan is to have adequate nutrition for the tree through ground application. Foliar sprays can sometimes be useful if a quick and short-lasting effect is needed. Parades *et al.* (1999) pointed out that OVW has a high potassium concentration and notable levels of nitrogen, phosphorus, calcium, magnesium, and iron. Hence, a conditioning treatment of this waste is necessary to produce a stable and easily management able end-product (Parades *et al.*, 2005).

Stone length, diameter, weight and oil content

The effect of the tested treatments on stone length, diameter and stone weight of *Manzanillo* olive trees is shown in Table 13. Differences were not significant with most of treatments. Despite that all treatments had no significant effect among both of soil drench application and foliar spray, in the first season , soil drench application gave the highest significant mean values of stone length (1.63 and 1.54 cm, respectively) at both seasons. The highest significant value of 1.64 cm was recorded by trees received 24 liter of Cyano/OVW+100% NPK in the second season. As for stone diameter, Application of NPK at the 50% of the control dosage + 48 liter Cyano/OVW recoded relatively higher stone diameter (1.02 cm) compared with other treatment in the first season, but in the second season there were no significant differences with regard to the specific effect.

Concerning the stone weight, results revealed that application of NPK at (50%) of the control dosage + 48 liter of Cyano/OVW gave the heaviest stone weight (1.08 g) in the first season while, in the second season the control had the highest values. As for fruit oil content, Table (13) indicated that soil drench and foliar spray application methods had no significant effect on oil content during the first season. However, the highest oil values at the first season (36.63 and 37.16%) recorded by trees received 75%NPK of the control dosage + Cyano/OVW soil drench application (36 L/tree) and others received 50% NPK of the control dosage + 50% Cyano/OVW foliar spray (48 L/tree), respectively. At the second season, the highest records of 39.18 and 38.75% were for applying

Cyano/OVW soil drench application (24 L/tree) + NPK at 100% of the control dosage and applying 50% NPK of the control dosage + 50% Cyano/OVW foliar spray (48 L/tree), respectively. These results are nearly in the same line with these obtained by Shaheen *et al.* (2011) on using olive vegetation water on growth, flowering and yield of *Manzanillo* olive trees.

TABLE 13. Effect of Cyano/OVW biofertilizer on stone length, diameter and weight as well as fruit oil content of Manzanillo olive trees during 2013 and 2014 seasons.

Treatments		Stone length (cm)		Stone diameter (cm)		Stone weight (g)		Fruit oil content %	
		2013	2014	2013	2014	2013	2014	2013	2014
Soil drench application	Control	1.60ab	1.57bc	0.98ab	0.99a	0.96b	1.02a	30.14e	32.08d
	100%	1.62a	1.64a	0.94bc	0.97a	0.84c	0.86bc	34.23b	39.18a
	75%	1.63a	1.43d	0.94bc	0.95a	0.83c	0.79d	36.63ab	38.30ab
	50%	1.63a	1.53c	1.02a	0.99a	1.08a	0.86bc	31.88d	33.90c
Mean		1.63A	1.54A	0.97A	0.98A	0.93A	0.88A	33.97A	35.86A
Foliar spray application	Control	1.60ab	1.57bc	0.98ab	0.99a	0.96b	1.02a	30.14e	32.08d
	100%	1.60ab	1.60ab	1.00a	0.99a	0.99b	0.91b	33.42c	25.08e
	75%	1.57ab	1.40d	0.89c	0.94a	0.79c	0.75d	35.60b	37.37b
	50%	1.50b	1.40d	0.89c	0.93a	0.79c	0.80cd	37.16a	38.75a
Mean		1.57B	1.49B	0.94A	0.96A	0.88B	0.87A	34.08A	33.32B
Mean	Control	1.60A	1.57B	0.98A	0.99A	0.96A	1.02A	30.14C	32.08C
	100%	1.61A	1.62A	0.97A	0.98AB	0.91B	0.88B	35.33AB	32.13C
	75%	1.60A	1.42D	0.92B	0.94B	0.81C	0.77D	36.12A	37.83A
	50%	1.58A	1.47C	0.96A	0.96AB	0.94AB	0.83C	34.52B	36.33B

Values within same column having different letters showed statistically significant differences ($p<0.005$)

Economic evaluation

Data presented in Table 14 and 15 show the effect of drench and foliar spray application of Cyano/OVW on net income of Manzanillo olive trees during 2013 and 2014seasons. Price/Kg averaged 3 EGP in 2013 and 2014.

TABLE 14. Economic evaluation of Cyano/OVW soil drench application on net return of olive trees during 2013 and 2014 seasons.

Treatments	Mean yield kg/fed		Gross income (EGP.fed^{-1})		Operation cost	Mineral Fertilizer cost	Cyano /OMV cost	Total cost	Net return (EGP.fed^{-1})	
	2013	2014	2013	2014	Average for two seasons (EGP.fed^{-1})				2013	2014
Control	945	1890	2835	5670	3204	2296	0	5500	-2665	170
100%	2415	3360	7245	10080	3204	2296	126	5626	1619	4454
75%	1050	2205	3150	6615	3204	1722	189	5115	-1965	1500
50%	1260	3150	3780	9450	3204	1148	252	4604	-824	4846

*Gross income (EGP.fed^{-1}) = Mean yield kg.fed^{-1} x Price sell of olive fruits (3.0 EGP/Kg)
*Operation cost (EGP.fed^{-1}) = Cost of irrigation, Labors, pesticides and others
* Price sell of Cyano + OVW = 0.1 EGP.L^{-1}
*Total cost (EGP.fed^{-1}) = Operation cost + Compost and mineral fertilizers cost + Cyano/OMV cost
*Net return (EGP.fed^{-1}) = Gross income - Total cost

TABLE 15. Effect of Cyano/OVW foliar spray application on net return of olive trees during 2013 and 2014 seasons.

Treatments	Mean yield kg.fed^{-1}		Gross income (EGP.fed^{-1})		Operation cost	Cyano /OMV	Total cost	Net return (EGP.fed^{-1})	
	2013	2014	2013	2014	Average for two seasons (EGP.fed^{-1})			2013	2014
Control	945	1890	2835	5670	5500	0	5500	-2665	170
100%	1050	3045	3150	9135	5500	10.5	5510.5	-2360.5	3624.5
75%	1785	3780	5355	11340	5500	10.5	5510.5	-155.5	5829.5
50%	2625	4305	7875	12915	5500	10.5	5510.5	2364.5	7404.5

*Gross income (EGP.fed^{-1}) = Mean yield kg.fed-1 x Price sell of olive fruits (3.0 EGP/Kg)
*Operation cost (EGP.fed^{-1}) = Cost of irrigation, Labors, pesticides and others
* Price sell of Cyano + OVW = 0.1 EGP.L^{-1}
*Total cost (EGP.fed^{-1}) = Operation cost + Compost and mineral fertilizers cost + Cyano/OMV cost
*Net return (EGP.fed^{-1}) = Gross income - Total cost

Total operation cost included cost of water, fertilizers, labors, pesticides and others. Soil drench application of Cyano/OVW (24 L/tree) plus 100% NPK achieved the highest net return (1619 EGP) for the first season while, Cyano/OVW (48 L/tree) plus 50%NPK recorded the highest net return (4846 EGP) for the second season. As for foliar spray application, 50% foliar spray technique with the full recommended NPK dose recorded the highest net return of 2364.5 and 7404.5 EGP for the first and second seasons, respectively and this treatment is the best economic recommended treatment.

Conclusion

The large proportion of organic matter and valuable nutrients make olive vegetative wastewater "OVW" a valuable resource for beneficial utilization, particularly in degraded agricultural soils of low organic matter content, abundant in the Mediterranean basin. In fact, it has been proven that this waste may potentially act as good sources of plant nutrients. In the present study, phycoremediation of OVW was conducted using mixture of cyanobacteria strains namely: *Nostoc muscorum*, *Anabaena oryzae* and *Spirulina platensis*. Using OVW as a substrate for growing N_2-fixing and PGPR-producing cyanobacteria was at the expense of its constituents and transform it into an organic liquid of high fertilizing value. Results indicated that Cyano/OVW had been successfully assayed as soil drench and foliar spray application fertilizer for improving the vegetative growth, fruit yield and increasing fruit oil content of *Manzanillo* olive trees and enhanced soil fertility. The end product "Cyano/OVW biofertilizer" was proved to be a natural liquid organic biofertilizer would be a sustainable economic OVW recycling option. Generally, it will be necessary to perform further field studies to know the effect of Cyano/OVW biofertilizer as biofertilizer on other plants and soils.

References

A.O.A.C. (1995) "*Official Methods of Analysis*", 15th ed., Association of Official Analytical Chemists.Washington, D.C., USA.

Abd El Monium, Eman, A. and Abd-allah, A. S. E. (2008) Effect of green Alga Extract as foliar Spray on vegetative growth, yield and berries quality of Superior grapevines. Amer-Euras. J. *Agric. & Environ. Sci.,* **4** (4), 427-433.

Abd El-Motty, Elham, Z., Shahin, M. F. M., El-Shiekh, M. H. and Abd-El-Migeed, M. M. M. (2010) Effect of Algae extract and yeast application on growth, nutritional status, yield and fruit quality of Keitte mango trees. *Agri. Biolo. J. N. Amer.,* **1** (13), 421-429.

Abou–Taleb, A. Safia, Abou El-Khashab, A. M. and Sead, T. Wafaa (2004) Growth and productivity of olive trees as affected by some bio and mineral nitrogen fertilizers. *Minufiya J. Agric. Res. Egypt.,* **29** (4), 933-963.

Abu-Zreig, M. and Al-Widyan, M. (2002) Influence of olive mills solid waste on soil hydraulic properties. Communications in Soil. *Sci. Plant Analysis,* **33**, 505-517.

Acea, M.J., Fernandez, A.P. and Cid, D.A. (2003) Cyanobacterial inoculation of heated soils: effect of microorganisms on C and N cycles and on chemical composition in soil surface. *Soil Biol. Biochem.,* **35**, 513-524.

Adam, M.S. (1999) The promotive effect of Cyanobacterium (Nostoc muscorum) on the growth of some crop plants. *Acta Microbiologica Polonica,* **48** (2), 163-171.

AL-Absi, K. M. (2010) Growth and biochemical of olives as influenced by irrigation with tread industrial of olive waste water. *J. plant Nutrition,* **33,** 1-14.

Alireza, T.M., Panahi, S.B. and Khezri, M. (2006) Effects of shoot girdling and urea combined with6- benzyladenine on abscission of inflorescence buds in "Ohadi" pistachio cultivar (Pistacia vera L.). *Int. J. Agric. Biol.,* **8** (4), 474-76.

Al-Sherif, E.A., Abd El-Hameed, M. S., Mervat, A. Mahmoud and Heba S. Ahmed (2015) Use of cyanobacteria and organic fertilizer mixture as soil bioremediation. *Amer.-Eurasian J. Agric. Environ. Sci.,* **15** (5), 794-799.

Aly, E.A. (2005) Physiological studies on the effect of salt stress on some olive cultivators. *Ph.D. Thesis,* Fac. Agric. Benha Univ., Egypt.

Amores-Sanchez, I., María del Carmen, T., Aldo, E.G. and Tania, G.V. (2015) Potential of microalgae and cyanobacteria in bioremediation of distillery wastewaters. ICIDCA. *Sobre los Derivados de la Caña de Azúcar,* **49** (1), 58-70.

APHA (1998) "*Standard Methods for the Examination of Water and Wastewater*", 20th ed., *Amer. Public Health Association Washington, DC.*

Araña, J., Tello Rendón, E., Doña Rodríguez, J., Herrera Melián, J., González Díaz, O. and Pérez Peña, J. (2001) Highly concentrated phenolic wastewater treatment by the Photo-Fenton reaction, mechanism study by FTIR-ATR. *Chemosphere,* **44** (10) 17–23.

Boraste, A., Vamsi, K., Jhadav, A., Khairnar, Y., Gupta, N., Trived, S., Patil, P., Gupta, G., Gupta, M., Mujapara, A. and Joshi, B. (2009) Biofertilizers: A novel tool for agriculture. Inter. *J. Microbiol. Res.*, **1**, 23-31.

Brown, J. D. and Lilleland, D. (1946) Rapid determination of potassium and sodium in plant material and soil extract by flame photometer. *Proc. Amer. Soc. Hort. Sci.*, **48,** p. 341.

Casida, L.E., Klein, D. A. and Santoro, T. (1964) Soil dehydrogenase activity. *Soil Sci.*, **98**, 371-376.

Chaari, L., Elloumi, N., Mseddi, S., Gargouri, K., Rouina, B. B., Mechichi, T. and Kallel, M. (2015) Changes in soil macronutrients after a long-term application of olive mill wastewater. *J. Agric. Chem. Environ.*, **4**, 1-13

Chapman, H.D. and Pratt, P.F. (1961) "*Methods of Analysis for Soils, Plants and Waters*". Div. of. Agr. Sci., Univ. Calif. U.S.A. 309 p.

Chartzoulakis, K., Psarras, G., Moutsopoulou, M. and Stefanoudaki, E. (2010) Application of olive wastewater to cretan olive orchard: Effects on soil properties, plant performance and the environment. *Agric. Ecosystem and Environ.*, **138**, 293-298.

Di Bene, C., Pellegrino, E., Debolini, M., Silvestri, N. and Bonari, E. (2013) Short- and long-term effects of olive mill wastewater land spreading on soil chemical and biological properties. *Soil Biology and Biochemistry*, **56**, 21-33.

Di Serio, M.G., Lanza, B., Mucciarella, M. R., Russi, F., Iannucci, E., Marfisi, P. and Madeo, A. (2008) International Biodeterioration and Biodegradation. **62**, 403-407.

Duarte, J.C., O. Pires, Susana, Paixão, S.M. and Céu Sàágua, M. (2011) New approaches to olive mill wastes bioremediation. *Olive Oil and Health, Chapter 11. Nova Sc. Publishers Inc., pp. 289-308.*

EI-Sheekh, M.M., Ghareib, M.M. and El-Souod, G.W.A. (2012) Biodegradation of phenolic and polycyclic aromatic compounds by some algae and cyanobacteria. *J. Bioremed Biodegrad*, **3,**133. doi: 10.4172/2155-6199.1000133

El- Karamany, M.F., Ahmed, M.K.A., Bahr, A.A. and Kabesh, M.O. (2000) Uilization of bio-fertilizers in field crop production. *Egypt J. Appl. Sci.*, **15**, 137.

El-Sharony, T.A.M. (2007) Effect of organic and bio- fertilization on growth and productivity of olive trees. *MSc. Thesis,* Fac. Agric. Cairo Univ., Egypt.

El-Taweel, A.A. (2005) Studies on fertilization of Guava trees. *Ph.D. Thesis,* Fac. Agric. El-Mansoura Univ., Egypt.

Fernandes, T.A., Iyer, V. and Apte, S.K. (1993) Differential responses of nitrogen fixing cyanobacteria to salinity and osmotic stress. Applied and Environ. *Microbiology*, **59**, 899-904.

Fernandez J. E., Moreno, F., Cabrerera, F., Arrue, J. L. and Martin-Aranda, J. (1991) Drip irrigation, soil characteristics and the root distribution and root activity of olive trees. *Plant and Soil.* **133**, 239-251.

Garcia, L. (2013) Short-term effects of olive mill by-products on soil organic carbon, total N, C/N ratio and stratification ratios in a Mediterranean olive grove. *Agriculture, Ecosystems & Environment.,* **165**, 68-73.

Grzesik, M. and Romanowska-Duda, Z. (2015) Ability of cyanobacteria and green algae to improve metabolic activity and development of willow plants. *Pol. J. Environ. Stud.,* **24** (3), 1003-1012.

Hachicha, S., Cegarra, J., Sellami, F., Hachicha, R., Drira, N., Medhioub, K. and Ammar, E. (2009) Elimination of polyphenols toxicity from olive mill wastewater sludge by its co-composting with sesame bark. *J. Hazardous Materials,* **161**, 1131-1139.

Hardy, R.W.E., Burns, R.C. and Holsten, R.D. (1973) Application of the acetylene-ethylene assay for measerment of nitrogn fixation. *Soil Biol. Biochem.,* **5**, 47-81.

Heady, E.O. and Dillon, J.L. (1961) Agricultural Production Functions. Iwoa State University press. Iwoa State University press, Iwoa, U.S.A.

Hegab, M.Y., Sharawy, A.M.A. and El-Saida, S.A.G. (2005) Effect of algae extract and mono potassium phosphate on growth and fruiting of Balady orange trees (Citrus sinensis). *Proc. First Sci. Conf. Agric. Sci. Fac. of Agric., Assuit Univ.,* **1**, 73-84.

Hegazi, E.S. and Stino, G.R. (1982) Chemical regulation of sex expression in certain olive *cultivars. Acta Agrobotanica,* **35** (2), 185-190.

Hegazi, E.S., El-Sonbaty, M.R., Eissa, M.A. and El-Sharony, T.F.A. (2007) Effect of organic and bio-fertilization on vegetative and flowering of Picual olive trees. *World J. Agric. Sci.,* **3**, 210-217.

Jackson, M.L. (1973) "*Soil Chemical Analysis"*. Prentice-Hall, Inc., N.J. Private Limited and New Delhi, India.

Jaouani, A., Vanthournhout, M. and Penninckx, M.J. (2005) Olive Oil Mill Wastewater Purification by Combination of Coagulation- Flocculation and Biological Treatments. *Environmental Technology,* **26**, 633-642.

Kavdir, Y. and Killi, D. (2008) Influence of olive oil solid waste applications on soil pH, electrical conductivity, soil nitrogen transformations, carbon content and aggregate stability. *Bioresource Technology,* **99**, 2326-2332

Kulk, M.M. (1995) The potential for using Cyanobacteria (blue-green algae) and algae in the biological control of plant pathogenic bacteria and fungi. *European J. Plant Pathol.,* **101**(6), 585-599.

Lia, B.Y., Zhoua, D. M., Canga, L., Zhangc, H. L., Fana, X. H. and Qina, S. W. (2007) Soil micronutrient availability to crops as affected by long-term inorganic and organic fertilizer applications. *Soil and Tillage Res.,* **96**, 166-173.

Magdich, S., Ben Ahmed, C. and Jarboui, R. (2013) Dose and frequency dependent effects of olive mill waste water treatment on the chemical and microbial properties of soil. *Chemosphere*, **93**, 1896-1903.

Magdich, S., Jarboui, R., Ben Rouina, B., Boukhris, M. and Ammar, E. (2012) A yearly spraying of olive mill wastewater on agricultural soil over six successive years: Impact of different application rates on olive production, phenolic compounds, phytotoxicity and microbial counts. *Sci. of the Total Environment*, **430**, 209-216.

Mahmoud, A.A., Soha S.M. Mostafa, Azza A.M. Abd El All and Hegazi, A.M. (2007) Effect of cyanobacterial inoculation in presence of organic and inorganic amendments on Carrot yield and sandy soil properties under drip irrigation regime. *Egypt. J. Appl. Sci.*, **22** (12B), 716-733.

Mahmoud, Y. I., Soha S.M. Mostafa and Faten M. Mohamed (2015) Comparative evaluation of biological treatments and mineral NPK on rice productivity in alkaline-saline Middle East *J. Agric. Res.*, **4** (4), 735-744.

Maksoud, M.A. (2000) Response of growth and flowering of Manzanillo olive trees to different sorts of nutrients. *Egypt. J. Hort.*, **27** (4), 513-523.

Mekki, A., Dhouib, A. and Sayadi, S. (2006) Changes in microbial and soil properties following amendment with treated and untreated olive mill wastewater. *Microbiological Res*., **161**, 93–101.

Mekki, A., Dhouib, A. and Sayadi, S. (2007) Polyphenols dynamics and phytotoxicity in a soil amended by olive mill wastewaters. *J. Environ Manage*, **84**, 134–140.

Mekki, A., Dhouib, A. and Sayadi, S. (2009) Evolution of several soil properties following amendment with olive mill wastewater. *Prog. Natl. Sci.*, **19**,1515–1521.

Mofeed, A.S. (2002) Effect of picking dates on flowering and fruiting in olive tree. Faculty of Agriculture, Cairo University. Egypt.

Mostafa, Soha S.M., Amal W. Abou Elkhair and Heba Sh. Shhata (2013) Effect of plant growth promoting substances from rhizo- and cyano-bacteria on sugar beet growth, yield and yield quality in saline soil. *Intr. J. Acad. Res*., **5** (6), 58-65.

Mostafa, Soha S.M., Eman A. Tantawy, Manal A.H. El-Gamal and Abd El-Halim, A.Kh. (2011) Effect of cyano- and endophytic – bacteria on wheat productivity under nitrogen limitation or irrigation regime in calcareous soil. *N. Egypt. J. Microbiol.*, **30**, 210-241.

Mostafa, Soha S. M., Mohamed, A. I. Shehata, Heba SH. and Awad, A. E. (2015) Enviro-economic impact of cyanobacteria, bacteria, vinasse and pretreated rice straw to secure yield productivity of rice crop in saline soils. *N. Egypt. J. Microbiol.*, **40**, 47-81.

Murphy, J. and Riely, J.P. (1962) A modified single dilution method for determination of phosphate in natural water. *Ann. Chemi. Acta*, **27**, 31-36.

Nanjappan, K., Radha, P., Lata, N. and Kaushik, B.D. (2007) Evaluating the potential of plant growth promoting cyanobacteria as inoculants for wheat. *European J. Soil Biology,* **43** (1), 23-30.

Osman, S.M., Khamis, M.A. and Thorya, A.M. (2010) Effect of Mineral and Bio-NPK soil application on Vegetative Growth, Flowering, Fruiting and leaf Chemical Composition of Young Olive Trees. Res. *J. Agric. Bio. Sci.,* **6** (1), 54-63.

Page, A.L., Miller, R.H. and Keeny, D.R. (1982) "*Methods of Soil Analysis*". Part 2. Chemical and Microbiological properties. 2nd ed., Madison, Wisconsin, USA.

Palaniappan, P., Malliga, P., Manian, S., Sivaramkrishnand, S., Madhaiyan, M. and Sa, T. (2010) Plant growth promontory effect on cow pea (Vigna unguiculata L.) using coir pith aqueous extract formulation of cyanobacterium Phormidium. *Amer. Eurasian J. & Environ. Sci.,* **8** (2), 178 -184.

Parades, C., Cegarra, J., Roig, A., Sfinchez Monedero, M.A. and Bernal, M.P. (1999) Characterization of olive mill wastewater (alpechin) and its sludge for agricultural purposes. *Bioresour. Tech,* **67**, 111-115.

Parades, C., Cegarra, J., Bernal, M.P. and Roig, A. (2005) Influence of olive mill wastewater in composting and impact of the compost on a Swiss chard crop and soil properties. *Environ. Inter*. **31**, 305-312.

Pimentel, M., Oturan, N., Dezotti, M. and Oturan, M.A. (2008) Phenol degradation by advanced electrochemical oxidation process electro-Fenton using a carbon felt cathode. *Appl. Catal. B Environ.,* **83**, 140–9.

Piotrowska, A., Rao, M. A., Scotti, R. and Gianfreda, L. (2011) Changes in soil chemical and biochemical properties following amendment with crude and dephenolized olive mill waste water (OVW). *Geoderma,* **16**, 8-17. http://dx.doi.org/10.1016/j.geoderma.2010.11.011

Piper, C.S. (1950) "*Soil and Plant Analysis*". Int. Sci. Publ., New York, 368 p.

Pregl, F. (1945) "*Quantitative Organic Micro-Analysis*", 4th ed., and A. Churchill. LTD. London. pp. 126-129.

Rana, A., Joshi, M., Prasanna, R., Shivay, Y.S. and Nain, L. (2012) Biofortification of wheat through inoculation of plant growth promoting rhizobacteria and cyanobacteria. European *J. Soil Biology,* **50**, 118–126. http://dx.doi.org/10.1016/j.ejsobi.2012.01.005

Sahraoui H., Kanzari, S., Hachicha, M. and Jmellouli, H. (2015) Olive mill wastewater spreading effects on hydraulic soil properties. *The Experiment,* **30** (4), 2002-2011

Sahu, D., Priyadarshani, I. and Rath B. (2012) Cyanobacteria - as potential biofertilizer. *CIBTech Journal of Microbiology,* ISSN: 2319-3867, 1 (2-3), pp.20-26.

SAS Institute (1994) "*SAS user's Guide: Statistics*". Version 6. 4th ed. SAA Inst., Cary, NT.

Shaheen, Shereen, A., El-Taweel, A.A. and Al- Khateeb, A. (2011) Effect of using olive vegetative water on growth, flowering and yield of Manzanillo olive trees. *J. Amer. Sci.,* **7** (9), 501-510.

Sherman, L., Min, H., Toepel, J. and Pakrasi, H. (2010) Better living through cyanothece - unicellular diazotrophic cyanobacteria with highly versatile metabolic systems. *Adv. Exp. Med. Biol.* **675**, 275-290.

Snedecor, G.W. and Cochran, W.G. (1980) "*Statistical Methods*", 7th ed., Iowa State Univ. Press, Ames, U.S.A. 507 p.

Ulger, S., Sonmez, S., Karkacier, M., Ertoy, N., Akdesir, O. and Aksu, M. (2004) Determination of endogenous hormones, sugars and mineral nutrition levels during the induction, initiation and differentiation stages and their effects on flower formation in olive. *Plant Growth Regulation,* **42** (1), 89-95.

Vossen, P.M. (2007) Site, Varieties, and production systems for organic olives. Chapter 1 in Organic Olive Production Manual. *University of Calif ornia Agriculture & Natural Resources; 1st edition (April 5, 2007). ISBN : 1601074409.*

Yoshida, T. and Ancajas, R. (1973) Nitrogen fixing activity in upland and flooded rice fields. *Soil Sci. Soc. Amer. Proc.*, **37**, 42-46.

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كفاءة استخدام السيانوبكتريا وماء خضري الزيتون في التسميد الحيوي لأشجار الزيتون تحت مستويات مختلفة من السماد المعدني

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أجريت هذه التجربة خلال موسمين متتاليين ٢٠١٣ و ٢٠١٤ على أشجار زيتون صنف منزانللو عمرها ١٣ سنة و النامية فى تربة رملية طفلية على مسافات ٥ × ٨ م باستخدام ممارسات زراعية موحدة تحت نظام الرى بالتنقيط فى مزرعة خاصة على بعد ٥٠ كم من القاهرة – طريق مصر الاسكندرية فى شمال غرب مصر لدراسة تأثيراستخدام السماد الحيوي الناتج من زراعة ناجحة لخليط ثلاث سلالات من السيانوبكتريا (نوستوك مسكورم، أنابينا أوريزا ، سيبروليا بلاتنسيس) على ماء خضري الزيتون المخفف بماء الصنبور (١:١) على بعض خصائص التربة والنمو والمحتوى المعدنى وخصائص الأزهار والمحصول وجودة ثمار أشجار الزيتون صنف المنزانللو. وأبرزت هذه الدراسة أيضا الجدوى الاقتصادية لهذه الممارسات في تحسين جودة الثمار وزيادة محصول الثمار، و ذلك من خلال استخدام طريقتين للتطبيق و هما الإضافة الأرضية و طريقة الرش على الأشجار. للإضافة الأرضية تم تخفيف السماد الحيوي الناتج بماء الصنبور (١:١) و أضيف للتربة بمعدلات ٢٤ ، ٣٦، ٤٨ لتر من السماد الحيوي للشجرة بمصاحبة ثلاث معدلات سمادية وهي جرعة كاملة من سماد المزرعة (١٠٠٪) من نتروجين وفوسفور وبوتاسيوم +٢٤ لتر ماء خضرى المعالج بالسيانوبكتريا و المخفف بماء الصنبور و(٧٥٪) من سماد المزرعة من نتروجين وفوسفور وبوتاسيوم +٣٦ لتر ماء خضرى زيتون المعالج بالسيانوبكتريا مخفف بماء الصنبور و(٥٠٪) من سماد المزرعة نتروجين وفوسفور وبوتاسيوم +٤٨ لتر ماء خضرى زيتون المعالج بالسيانوبكتريا مخفف بماء الصنبور. أما بالنسبة لتطبيق معاملات الرش الورقي، تم تخفيف حجم ثابت من الماء الخضري المعالج السيانوبكتريا (١ لتر/شجرة) مع ماء الصنبور (١: ٥) و كانت المعدلات ١٠٠٪من جرعة ماء خضرى الزيتون المعالج بالسيانوبكتريا مخففة بماء الصنبور يرش فى ٤ اتجاهات + الجرعة كاملة (١٠٠٪) من سماد المزرعة نتروجين وفوسفور وبوتاسيوم مضافة ارضياً، والمعاملة الثانية كانت الرش بمعدل ٧٥٪ (ثلاث اتجاهات) من جرعة ماء خضرى الزيتون المعالج بالسيانوبكتريا المخففة بماء الصنبور+ الجرعة الكاملة (١٠٠٪) من سماد المزرعة نتروجين وفوسفور وبوتاسيوم مضافة ارضياً، والمعاملة الثالثة كانت الرش بمعدل ٥٠٪ (اتجاهين المعرضين للشمس) من جرعة ماء خضرى الزيتون المعالج بالسيانوبكتريا المخففة بماء الصنبور + الجرعة (١٠٠٪) من نتروجين وفوسفور وبوتاسيوم مضافة أرضياً.

يمكن القول بصفة عامة أن المعاملات أدت إلى تحسن بعض صفات التربة الكيميائية والبيولوجية والنمو النباتى والمحصول وجودة الثمار وبعض الصفات الفسيولوجية والعناصر المعدنية في الأوراق في مقابل انخفاض محتوى الفينولات في كل من التربة و الأوراق بالمقارنة بالكنترول. وكانت معاملة الرش بمعدل ٥٠٪ (اتجاهين) من جرعة ماء خضرى الزيتون المعالج بالسيانوبكتريا المخففة بماء الصنبور+ الجرعة الكاملة (١٠٠٪) من نتروجين وفوسفوروبوتاسيوم إضافة أرضية الأعلى فى قيم النمو الخضرى وخصائص التزهير والمحصول والمضافة خلال ستة أشهر من يناير الى يونيو مرتين كل شهر .وهذه هى المعاملة التي يمكن أن نوصى بها تحت ظروف هذه الدراسة والظروف المشابهة وهذا ما أكدته دراسة الجدوى الاقتصادية.