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Growth and Mineral Accumulation in *Olea europaea* L. Trees Irrigated with Municipal Effluent

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Abstract: Effect of municipal effluent on survival, growth and mineral nutrient in *Olea europaea* L. trees were studied. A study was carried out at Tehran (Iran) in which olive trees were irrigated with municipal effluent and well water for a period of 7 years. We analyzed the soil; leaves and fruits of olive trees irrigated with municipal effluent and compared them with olive trees irrigated with well water. Observation included tree height, collar diameter, survival and plant mineral composition, mineral uptake and change in soil properties at 7 years of plant age. Application of municipal effluent produced better growth in *Olea europaea* L. trees. The trees irrigated with municipal effluent attained 3.26±0.07 m height, 13.35±0.41 cm collar diameter, but trees irrigated with well water attained 2.47±0.12 m height, 9.5±0.64 cm collar diameter at the age of 7 years. Irrigation with municipal effluent increases pH, EC, OM, NH_4-N, NO_3-N and PO_4-P in soil. Concentration of N, P, K, Ca and Mg were greater in leaves and fruits of trees irrigated with municipal effluent than those of the leaves of trees irrigated with well water. Municipal effluent may be useful in tree irrigation to increase biomass productivity. Further, reduction of toxic concentration of metal ions in effluents may be helpful for a long-term field application.

Key words: Growth, irrigation, municipal effluent, plantation, *Olea europaea* L.

INTRODUCTION

In suburban areas, the use of industrial or municipal wastewater is common practice in many parts of the world (Feigin *et al*., 1991; Sharma *et al*., 2007). The prudent use of limited water supplies is critical for improving the socio-economic conditions of fragile arid areas (Government of India, 2001). Increasing industrialization and urbanization in major metropolitan cities in dry areas leads to an increase in the availability and volume of wastewater. This wastewater can be used for the restoration of degraded land and the growth of vegetation having commercial and environmental value (Dighton and Jones, 1991). In addition, wastewater is a valuable source of plant nutrients and organic matter needed for maintaining fertility and productivity levels of the soil (Weber *et al*., 1996). The wastewater will have a different fate than being pumped into a river, agricultural crops can make use of the extra water and nutrients (Toze, 2006) and groundwater recharge is yet another positive outcome of wastewater irrigation. Wastewater is recognized to have direct effect on soil chemical properties. It affects supply of mineral macro and micro nutrients for plant growth, soil pH, soil buffer capacity and soil CEC (Munir *et al*., 2007). The use of wastewater in growing woodlots is a viable option for the economic disposal of wastewater. Additional benefits include biomass production and sequestration of excess minerals in the plant system (Neilson *et al*., 1989). In reality, municipal effluent other than providing water resources for the plantations is an enormous nutrient source

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(Meli *et al*., 2002; Rattan *et al*., 2005). Management of wastewater irrigation should consider the wastewater nutrient content, specific crop nutrient requirements, soil nutrient content and other soil fertility parameters (Mohammad and Ayadi, 2004). Therefore, decision about the application of effluent should be made based on the views of specialties of water, soil, plant and environment of each location (Naghshinehpour, 1998). *Olea europaea* L. is an important multipurpose tree species with the potential to provide fruit and soil recapitulation. The use of municipal effluent in growing *Olea europaea* L. in suburban areas could be beneficial for the production of fruit and would have aesthetic and environmental benefits as well. The objective of this study was to quantify the growth of *Olea europaea* L. trees and the mineral accumulation in plants and surrounding soil under municipal effluent application.

MATERIALS AND METHODS

The field study took place in the urban fringe area of Tehran city (5 km South of Tehran), (Latitude 35.37' N, Longitude 51.31' E, elevation of 1005 a.s.l.). The area has a semi-arid with mile-cold winters conditions and vital growing season (dry season) of seven months (Mid April-Mid November) (Fig. 1). The mean annual rainfall is 232 mm, with maximum falls during winter. The highest precipitation falls in March and the lowest in August. Maximum and minimum mean monthly temperatures are 31.01°C in August and 5.49°C in January and average annual temperature is 13.3°C. The warmest month occurs in August and the coldest in January. The soil is clay (according to US soil taxonomy) with 45% clay, 30% sand and 25% silt. The specie selected for this study was *Olea europaea* L.

In this study, two even-aged (7 years) olive forestations have been selected. The first stand (20000 m^2) was being irrigated by municipal effluent and the second (10000 m^2) by well water. The irrigations were carried on 8 day durations for 8 months/year (during April-November).

Municipal effluent and well water were sampled daily for 7 days and each day 3 samples (at 3 h intervals 7 am, 13 pm and 19 pm) during the 4 week of May, July, August, September, October and November 2005 to make a composite sample of each day. This collection provided 21 samples of municipal effluent in each month. Thus, there were 252 samples, which include 126 samples of effluent and 126 samples of well water. Three replicate polyethylene bottles (acid-washed) of capacity 100 mL were immersed one by one at an interval of 15 sec in an open drain that was being used for irrigation purposes and immediately after filling, 1 mL of concentrated HNO_3 was added to the water to avoid microbial utilization of heavy metals. These samples were brought to the laboratory, filtered through Whatman No. 42 filter paper and stored at 4°C (OMA, 1990). Each sample was analyzed for pH, Electrical Conductivity (EC) and concentration of macro and

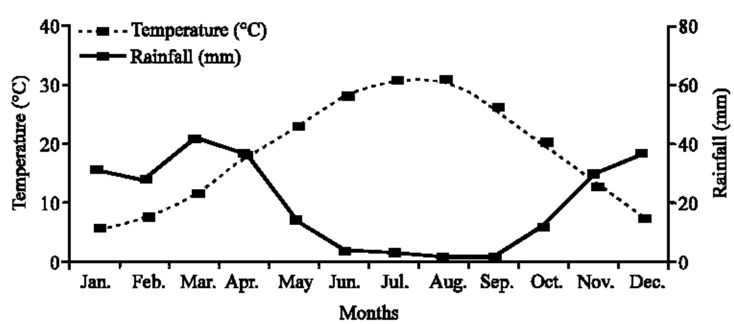

Fig. 1: Embrothermique curve of the study area

Table 1: Main characteristics of municipal effluent and well water (mean±SE)

Quality parameters	Municipal effluent	Well water	EPA USA Hach (2002)	WHO Hach (2002)	FAO (2000)
pH	7.63±0.01[a]	7.32±0.05[b]	6.5-8.5	6.5-8.5	6.5-8.5[a]
EC (dS m^{-1})	1.91±0.02[a]	0.59±0.008[b]	3	3	3
K (mg L^{-1})	39.93±0.83[a]	19.72±0.36[b]	---	---	---
Ca (g L^{-1})	255.22±4.57[a]	96.77±1.26[b]	75	400	400
Mg (g L^{-1})	109.85±1.83[a]	35.20±0.79[b]	50	50	60
NH_4-N (mg L^{-1})	9.05±0.11[a]	2.15±0.19[b]	---	5	---
NO_3-N (mg L^{-1})	1.63±0.09[a]	0.24±0.08[b]	---	3	---
PO_4-P (mg L^{-1})	12.69±0.16[a]	5.03±0.01[b]	---	---	---

Different superscripts in column are significantly different ($p<0.01$) (t-test)

micronutrients using standard procedures (OMA, 1990). Total alkalinity was determined by the titration method of OMA (1990). Nitrogen and phosphorus were analyzed calorimetrically (Jackson, 1973). Total parameters shown in Table 1, of municipal effluent and well water.

For the field measurements, four plots of 20×20 m were randomly identified in either of both areas and trees were measured in October for height, collar diameter, crown length, crown diameter and survival present.

Soil samples were collected in July 2005. In each plot, soil samples were taken from the root zone of each tree at three depths (0-15, 15-30 and 30-60 cm), by using a spiral auger of 2.5 cm diameter. Three sub-samples around the trunk were taken to make a composite soil sample per tree (Habibi Kaseb, 1992). They were transported to the laboratory, oven-dried at 40°C and crushed to pass through a 2 mm sieve. Soil pH (1:2.5 soil-water ratios), Electrical Conductivity (EC) and soil Organic Matter (OM) were determined using standard procedures (Jackson, 1973). For determination of macro and micronutrients, soil samples were extracted with ammonium acetate and sodium salt of Di-ethylene Tri-amine Penta-acetic Acid (DTPA) solution. Calcium (Ca) and magnesium (Mg) were estimated in absorption mode and potassium (K) in emission mode using a double beam atomic absorption spectrophotometer (model-3110, Perkin-Elmer, Boesch, Huenenberg, Switzerland). Available nitrogen was determined after 2 M KCl extraction. Extractable phosphorus was determined by the Olson's extraction method (Jackson, 1973).

In each plot, five olive individuals, growing in the terraces, were chosen and marked for sampling. Samples of the leaves and fruits were taken from trees, during the August and September. Samples were washed (for 10 sec approximately) with a solution of phosphate-free detergent, then with a 0.1 N HCl solution and finally with distilled water. Plant samples were wet digested as per Jackson (1973). Olive fruits (only the fleshy pulp) were also used to monitor nutrition elements. These were dried at 70°C, ground and passed through a 500 μm Stainless-steel sieve. Calcium (Ca) and magnesium (Mg) were estimated in absorption mode and potassium (K) in emission mode using a double beam atomic absorption spectrophotometer (model-3110, Perkin-Elmer, Boesch, Huenenberg, Switzerland). Measurement of N and P content were performed after a wet digestion using UV-vis spectrophotometer (Systronix model 117) at 490 and 420 nm.

Data were statistically analyzed using an SPSS package (Lindaman, 1992). Initially, normal distribution of the data was performed using the Shapiro-Wik's test. The difference between the concentrations of nutrient elements in the municipal effluent and well water, soils, leaves and fruits in the treatment and control sit were assessed by the independent-sample t-test. All statistical analyses were carried out with the program SPSS 12.5 for Windows.

RESULTS AND DISCUSSION

The results of current study conducted by the independent-sample t-test clearly showed that pH, EC (Electrical Conductivity) K, Ca, Mg, NH_4-N, NO_3-N and PO_4 -P, in municipal effluent were significantly greater ($p<0.01$) than well water (Table 1).

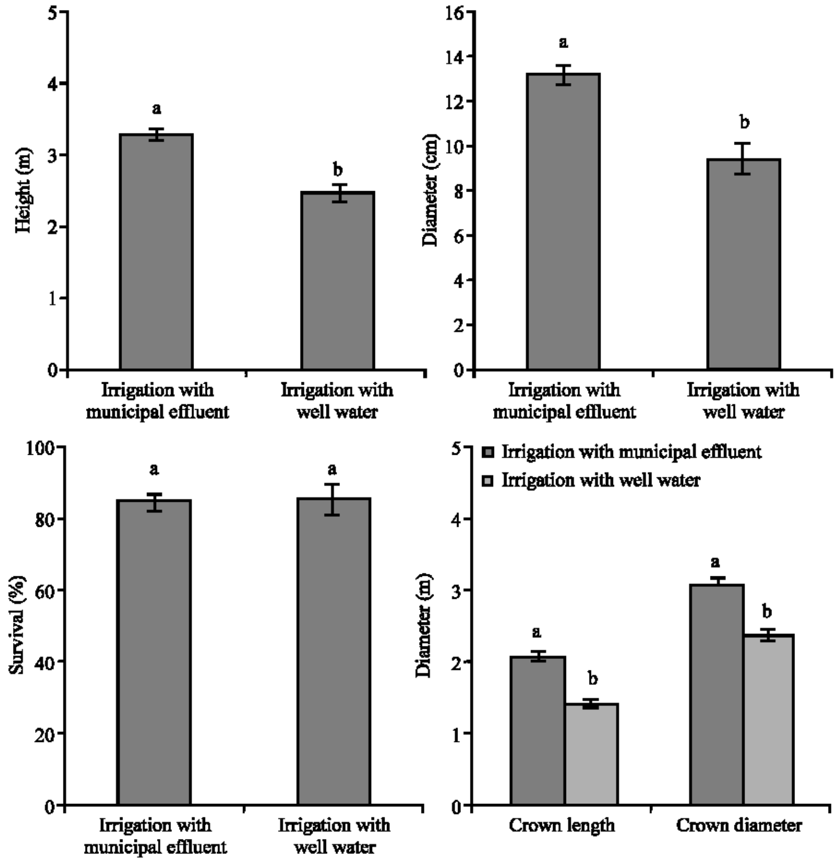

Fig. 2: Comparison of parameters measured of *Olea europaea* L. trees in two irrigated areas different superscripts in figure are significantly different ($p<0.01$) (t-test)

The survival of trees did not differ ($p>0.05$) among the treatments. Irrigation with municipal effluent for 7 years produced the largest trees (3.26±0.07 m height, 13.35±0.41 cm collar diameter, 3.11±0.04 m crown diameter and 2.11±0.06 m crown length) in treatment municipal effluent ($p<0.01$). The Survival, collar diameter, height, crown diameter and crown length for the trees of irrigated with municipal effluent were -0.76, 40.52, 31.98, 28.51 and 45.51%, respectively, greater than that of the trees irrigated with well water (Fig. 2).

Independent sample t-test indicated that a seven-year application of municipal effluent resulted increase (0-60 cm soil layers; mean of soil layers and treatments) in soil pH, EC, concentration of organic matter (OM), K, Ca, Mg, NH_4-N, NO_3-N and PO_4-P when compared to their values with well water ($p<0.01$, Table 2).

Mineral element concentrations in trees differed significantly ($p<0.01$) both due to the trees parts and treatments. Leaves had the highest concentration of nutrients followed by fruit in all the two treatments under study. N, P, K, Ca and Mg concentrations were high in the trees irrigated with municipal effluent followed by well water (Fig. 3 and 4).

Application of municipal effluent had a positive influence on the growth and production of *Olea europaea* L. survival extension of the trees irrigated with well water may be due to decreased loading of heavy metal. Accumulation of salts and some nutrients and heavy metals, after 7 years of municipal effluent irrigation, could have caused such reduction in the trees survival (Singh and Bahati,

Table 2: Effect of 7 years application of municipal effluent on physicochemical properties of soil

Quality parameters	Soil depth 0-15		15-30		30-60	
	T_1	T_2	T_1	T_2	T_1	T_2
Tissue	Clay	Clay	Clay	Clay	Clay	Clay
pH	7.95±0.016^{a}	7.68±0.032^{b}	7.93±0.075^{a}	7.67±0.017^{b}	7.95±0.014^{a}	7.71±0.017^{b}
EC (dS m^{-1})	0.89±0.05^{a}	0.69±0.07^{b}	0.85±0.068^{a}	0.64±0.008^{b}	0.81±0.085^{a}	0.63±0.008^{b}
OM (%)	0.83±0.13^{a}	0.37±0.045^{b}	0.41±0.038^{a}	0.24±0.045^{b}	0.29±0.06^{a}	0.11±0.03^{b}
K (g kg^{-1})	3.45±0.029^{a}	2.97±0.015^{b}	3.11±0.008^{a}	2.53±0.018^{b}	3.02±0.012^{a}	2.11±0.012^{b}
Ca (g kg^{-1})	23.63±1.21^{a}	15.96±1.29^{b}	21.63±0.93^{a}	15.03±0.95^{b}	21.33±0.77^{a}	15.96±1.29^{b}
Mg (g kg^{-1})	0.39±0.004^{a}	0.34±0.003^{b}	0.37±0.001^{a}	0.32±0.001^{b}	0.34±0.002^{a}	0.32±0.008^{b}
NH_4-N (mg kg^{-1})	44.60±1.32^{a}	27.17±1.01^{b}	39.90±1.00^{a}	21.72±1.35^{b}	28.14±1.02^{a}	18.33±1.24^{b}
NO_3-N (mg kg^{-1})	1.92±0.021^{a}	1.48±0.024^{b}	2.15±0.025^{a}	2.08±0.034^{b}	3.15±0.013^{a}	3.05±0.022^{b}
PO_4-P (mg kg^{-1})	31.52±1.02^{a}	25.67±1.21^{b}	29.30±1.03^{a}	19.74±1.30^{b}	14.14±1.22^{a}	11.73±1.20^{b}

T_1: Irrigation of seedlings with municipal effluent; T_2: Irrigation of seedlings with well water. Different superscripts in column are significantly different (p<0.01) (t-test), Values are mean of three replications±SE

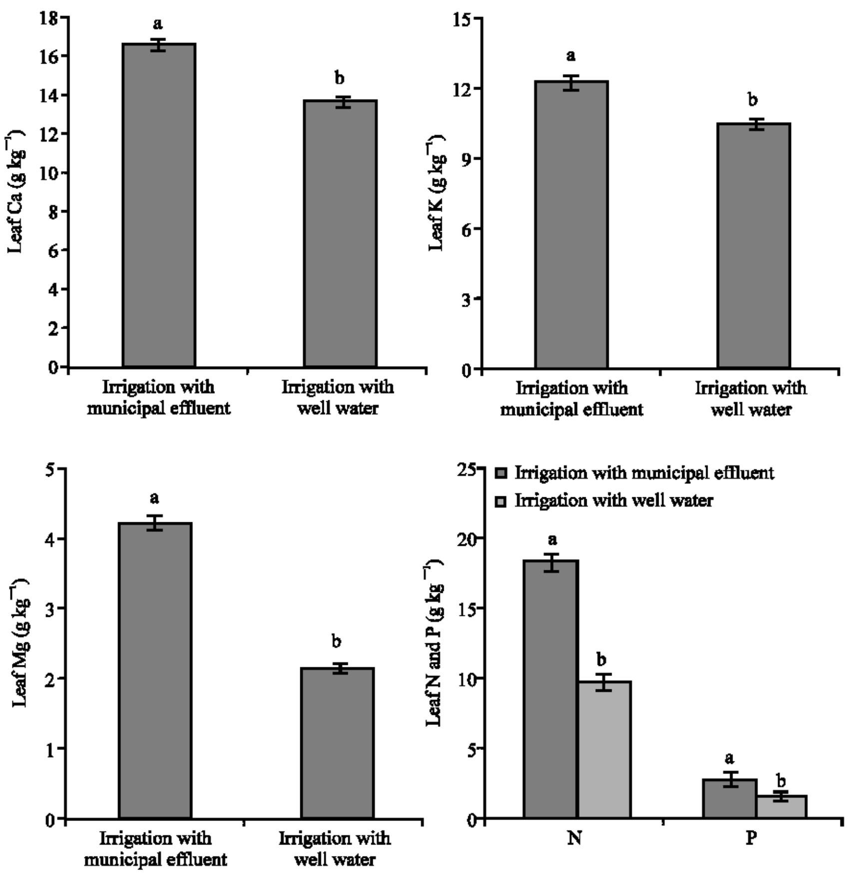

Fig. 3: Comparison of nutrient elements measured of *Olea europaea* L. leaf in two irrigated areas different superscripts in figure are significantly different (p<0.01) (t-test)

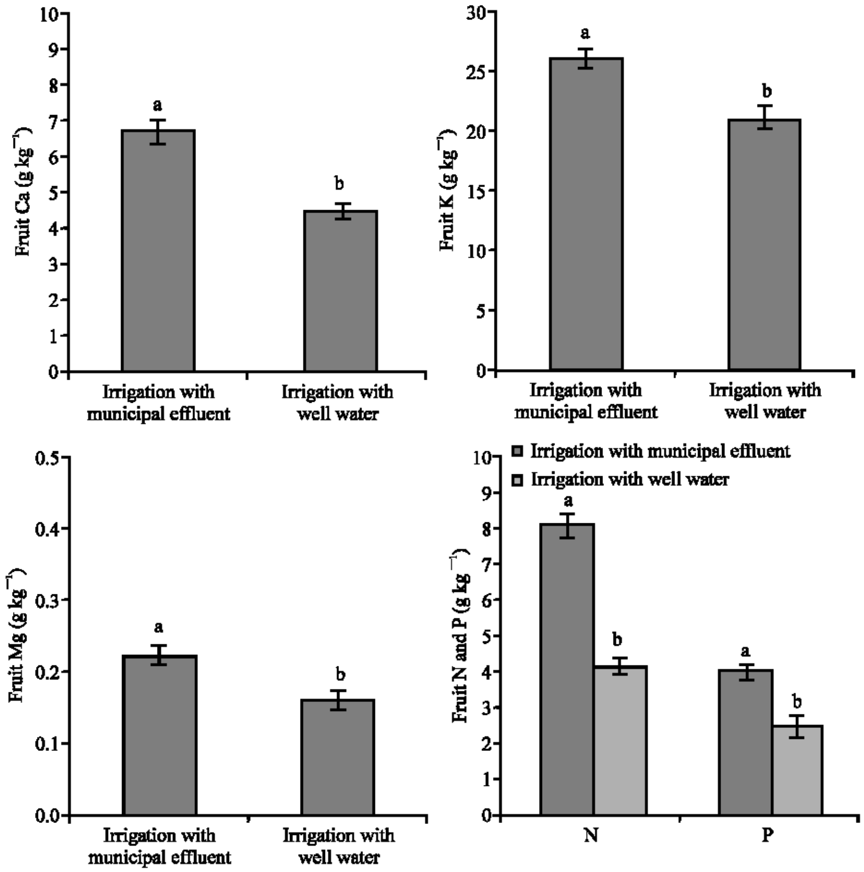

Fig. 4: Comparison of nutrient elements measured of *Olea europaea* L. fruit in two irrigated areas different superscripts in figure are significantly different ($p<0.01$) (t-test)

2005). Municipal effluent resulted in stimulation of growth and biomass production. Greater growth and biomass production of the trees irrigated with municipal effluent may be due to sufficient availability of water and essential elements (Guo *et al*., 2006). High growth in the trees irrigated with municipal effluent is obviously due to addition of nitrogen and phosphorus through municipal effluent addition. The increase in all growth parameters suggested that applied treatment had influenced the physiological processes, facilitated early leaf initiation and resulted in a net increase in the number of leaves. Increasing in leaves can captured more solar energy for metabolic use, fixed more CO_2 and produced greater photosynthetic and growth. This hypothesis is supported by Ceulemans *et al*. (1993) and Myers *et al*. (1996). Addition of municipal effluent on *Eucalyptus grandis* has been noted to result in a doubling of growth rate when compared to *E. grandis* grown in a rain fed site in four years (Stewart *et al*., 1990). Rigueiro Rodriguez *et al*. (2000) stated that the increase in the height and diameter of pine trunks, growing in plots irrigated with municipal effluent was significantly higher than that of trees growing in control and inorganically fertilized plots. An increase in the stem diameter and above ground biomass of a *Pinus radiata* D. Don due to fertilization has also been reported by Sheriff *et al*. (1986). The study of Qgbonnaya and Kinako (1993) also suggested that the seedlings supplied with near optimal amounts of water and nutrient or sewage water had greater growth rates than non-irrigated, unfertilized control seedlings of *Eucalyptus globules*. Both added municipal effluent and nutrients provided with wastewater application can be attributed to such increase in growth and production of *Olea europaea* L. Similar results were reported by Stewart and Flinn (1984), on *Pinus eldarica*, Ostos *et al.* (2007) on *Pistacia lentiscus* who observed that faster growth occurs in the effluent-irrigated areas. They attributed this increase to the organic matter and macro and micronutrient concentrations in the added wastewater. The wastewater contains considerable amount of nitrate, phosphate and potassium which are considered essential nutrients for improving plant growth and soil fertility.

The inconsistency in municipal effluent irrigation effect on soil pH was reported by other researchers. Schipper *et al.* (1996) found that soil pH increased following long term wastewater irrigation and they attributed this increase to the chemistry and high content of Na, Ca and Mg in the wastewater applied for a long period. The increase in the soil pH and EC may have been due to alkaline nature of and salt concentration in effluent (Mitra and Gupta, 1999). The soil pH affected with the pH of the wastewater (Parizek *et al.*, 2006). The increase in pH is common for soils receiving wastewater irrigation (Hu *et al.*, 2006) and can help reduce farming costs because the soils do not require lime. Mohammad and Mazahreh (2003) stated that the increase in EC for soil irrigated with wastewater compared with soil irrigated with potable water attributed to the original high level of (TDS) of the wastewater. Soil Organic Matter (OM) significantly increased with municipal effluent irrigation application, which is attributed directly to the contents of the nutrients and organic compounds in the municipal effluent applied. The soil OM contents accumulated more in the topsoil in all treatments. Similar increases in organic matter content have also been noted by Walker and Lin (2007) and Walker (2006) on soil irrigated with wastewater. Vazquezmontiel *et al.* (1996) found no positive effect on soil organic matter content with wastewater irrigation, while other researchers reported an increase in the soil organic matter following wastewater irrigation (Mancino and Pepper, 1992). Where more was accumulated in the topsoil (Mohammad and Mazahreh, 2003). Increase in OM content might be due to municipal effluent application (Kumar *et al.*, 1998). Baddesha *et al.* (1985) also suggested an increase in OM may be due to municipal effluent application. The availability of NH_4-N, NO_3-N, PO_4-P, K, Ca and Mg were greater in the soil irrigated with municipal effluent as compared to the soil irrigated with well water, suggesting the ameliorative effect of the species through absorption and accumulation in trees parts (Singh and Bahati, 2005) and due to their addition through municipal effluent in spite of their high uptake by the growing plants. Baddesha *et al.* (1985) have also reported that irrigation of soil under Eucalyptus plantation with sewage water increased the macro and micronutrient concentration. Municipal effluent irrigation increased significantly K, Ca, Mg, NH_4-N and PO_4-P in the top soil (0-15 cm, Table 2). Several researchers reported accumulation of N, P and K in the soil with wastewater application which was attributed to the original contents of these nutrients in the wastewater applied (Monnett *et al.*, 1996). These results agree with those reported by Day *et al.* (1979) and Mohammad and Mazahreh (2003) who found that extractable phosphorus was higher in soils irrigated with wastewater than in soil irrigated with fresh water or rainfall water. Nyamangara and Mzezewa (2000) found in their experiment that the concentration of K on the control treatment was decreased within depth and this increase in the soil surface was attributed to their high content in the wastewater used (Mohammad and Mazahreh, 2003). Considering the variation in the chemical composition of the wastewater, management of wastewater reuse should account for the N, P and K content prior to determination of the rate of wastewater application. Other researchers found that wastewater irrigations increased soil nitrogen N, phosphorus P and potassium K, while heavy metal levels tended to generally increase in soil with increasing number of years of irrigation (Vazquezmontiel *et al.*, 1996; Jalali *et al.*, 2007).

The discrepancy in nutrient concentration and uptake in *Olea europaea* L. may arise due to water and nutrient supply. Plant essential nutrient (total N, P and K) were higher in plants grown in soils irrigated with municipal effluent.

Enhancement of plant N content with wastewater application indicates that municipal effluent application provided the soil with these nutrients which enhanced required for plant growth and soil fertility. Nitrogen concentration in plant shoots was reported to be higher when grown with wastewater (Day *et al*., 1979), who found that N recovery in plants with wastewater was higher than the N recovery in plant material grown with well water. These results were attributed to significant increase in soil nitrogen with municipal effluent irrigation compared with the control. On the other hand, Papadopoulos and Stylianou (1988) reported that during the third irrigation season for trickle irrigation cotton (*Gossypium hirsutum* L. cv.); the N in petioles was greater with the treated effluent supplemented with no nitrogen, also in lamina. Phosphorus concentration were higher in plants grown in soils irrigated with municipal effluent There was significant ($p<0.01$) difference in phosphorus concentration between the control and the 7 years irrigated with municipal effluent. K, Ca and Mg concentration in leaves and fruits were increased significantly in the municipal effluent treatment. Similar results were reported by Herpin *et al.* (2007) on *Coffea arabica* L. Other researchers have reported an increase in P and K, Ca and Mg uptake by the plants irrigated with treated wastewater (Papadopoulos and Stylianou, 1988; Mohammad and Mazahreh, 2003; Singh and Bahati, 2005).

Soil and trees quality parameters are significantly affected by 7 years municipal effluent irrigation. This is mainly determined by the management of wastewater irrigation and its composition. In addition, continuous irrigation with municipal effluent may lead to accumulation of salts, plant nutrients and heavy metals beyond crop tolerance levels. Therefore, these concerns should be essential components of any management of municipal effluent irrigation. On the other hand, plant growth, soil fertility and productivity can be enhanced with properly managed municipal effluent irrigation, through increasing levels of plant nutrients and soil organic matter. It can be concluded, based on these results that proper management of municipal effluent irrigation and periodic monitoring of soil fertility and quality parameters are required to ensure successful, safe and long term reuse of wastewater for irrigation. Trees irrigated with municipal effluent showed optimum plant nutrient concentration. But use of municipal effluent in long term may be increase heavy metal in soil and plant tissues. Moreover, in general, distribution of municipal-treated wastewater for irrigation over large land areas causes minimal pollution hazard. It may be used as an optimal strategy for raising woodlot to supply fuel wood in vicinity of a suburban area.

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