

Effects of treated wastewater irrigation on lemon trees

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Received 24 April 2008; Accepted 29 July 2008

Abstract

This research is focused on evaluating the effects of applying treated wastewater on citrus trees. Two experimental plots irrigated with two different treated wastewater effluents were compared. The experimental sites were located in Murcia, southeastern Spain. The first experimental plot was located in Cartagena, where the treated wastewater had received a secondary treatment. The second experimental plot was located in Campotejar; in this case the water used was a mix of well water and wastewater from a tertiary treatment plant. The electrical conductivity (EC), turbidity and total dissolved solids (TDS) were higher in Cartagena's treated wastewater than in Campotejar's. Therefore, the mix with well water improved the agronomic quality of the reclaimed wastewater. The high levels of EC observed in both locations were due mainly to high chloride and boron (B) concentrations. Although leaf toxicity levels were not observed, the high salinity and B accumulation can be considered the main problems for the irrigation with treated wastewater in the region of Murcia. Microbiological analysis revealed an absence of faecal coliforms, *E. coli* and helminth eggs in the treated wastewaters and soil of Campotejar, but in Cartagena's treated wastewater faecal coliforms exceeded health standards.

Keywords: Treated wastewater reuse; Irrigation; Plant and soil effects

1. Introduction

In many parts of the world, treated wastewater has been successfully used for irrigation, and many researchers have recognized its benefits [1,2]. In the Mediterranean countries, treated

wastewater is increasingly used in areas with water scarcity and its application in agriculture is becoming an important addition to water supplies. In Greece the possibility of wastewater reuse for irrigation of vegetables has been studied by Kalavrouziotis et al [3]. They concluded that the future perspectives favour such a reuse, but to

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Presented at the 2nd International Congress, SMALLWAT 07, Wastewater Treatment in Small Communities, Seville, Spain, 11–15 November 2007.

accomplish social acceptance, more work is necessary to decrease the health risk factor involved and make the reuse safer.

Several studies have shown the advantages and disadvantages of using wastewater for irrigation of various crops [4]. The reuse of treated wastewater is a good option for increasing water supplies to agriculture. One of its benefits is the plant's use of the water's nutrients and therefore a reduction in the pollution load that wastewater contributes to the surface water supply [5]. However, depending upon its sources and treatments, sewage wastewater may contain high concentrations of salts, heavy metals, viruses and/or bacteria and the reclaimed wastewater application may create undesirable effects in soils and plants with direct effects on soil suitability for cultivation and water resources availability.

Current water quality criteria for agricultural reuse have mainly focused on total dissolved solids (TDS), salinity aspects [6], and the microbiological factors that may cause sanitary problems [7]. More specific water quality parameters for the reuse of reclaimed wastewater have been presented by Levine and Asano [2], and there is a considerable interest in the long-term effects of reclaimed wastewater on crops intended for human consumption.

The purpose of the present work was to study the effects of treated municipal wastewater reuse for the irrigation of citrus trees. The objective of this research was to compare two sources of treated wastewater, one obtained with a secondary treatment and the other with a tertiary treatment, and to study their effects on soil chemical properties and on the leaf mineral status.

2. Methodology

2.1. Experimental conditions

The experiment was conducted during 2006 in two locations in the region of Murcia. Carta-

gena's experimental plot is located in San Felix, a small village 4 km to the north of Cartagena (37°3'N, 0°58'W). The orchard size is 12 ha with a Fino lemon tree grafted on *Macrophyla* rootstock. The trees are 7 years old and the plant spacing 7×5 m. The water is supplied by drip irrigation with eight compensated pressure drips per tree, each with a flow rate of 4 Lh⁻¹. Campotejar's experimental plot is located 7 km to the north of Molina de Segura (38°07'N, 1°13'W). In this case the orchard size is 10 ha, cultivated with the same crop (variety and rootstock), tree age, and plant spacing and irrigation management as in Cartagena.

The soil grain size analyses were carried out by laser diffraction. According to the texture-triangle of the US Department of Agriculture, the soil was classified as silty loam in Cartagena and silty clay in Campotejar. The average annual precipitation in Cartagena and Campotejar ranged between 200 and 300 mm respectively. The average annual temperature in Campotejar was 19.7°C and in Cartagena was 18.7°C. During the season of the experiment, the mean daily reference evapotranspiration in Cartagena was 3.87 mm and in Campotejar 3.41 mm. Therefore, the greatest difference between both experimental plots was the origin and quality of the water used for irrigation. In Cartagena, wastewater with a secondary treatment was used, while in Campotejar the wastewater was treated with a tertiary treatment and mixed with well water in equal proportions.

The waste water irrigation was applied daily from January 2006 until December 2006, satisfying crop irrigation requirements determined according to daily crop reference evapotranspiration (ET_o), calculated with the Penman–Monteith equation [8], a crop factor based on the time of the year (FAO 56) and the percentage of ground area shaded by the tree canopy [9]. Micrometeorological data were collected by an automatic weather station located near the experimental site. Total water amounts applied to both

locations were measured within inline water meters and were 550 and 510 mm, in Cartagena and Campotejar, respectively.

2.2. Water analysis

Water samples were collected three times throughout 2006 (May, September, and December) in order to characterize irrigation water quality in both locations. Four samples for each irrigation source and time were transported on an ice chest with ice to the lab, stored and processed. The concentration of macronutrients (Na, K, Ca, Mg), micronutrients (Fe, B, Mn) and heavy metals (Ni, Cd, Cr, Cu, Pb, Zn) were determined by inductively coupled plasma atomic emission spectrometry (ICP–AES, Interprid II XDL); anions (chlorides, nitrates, phosphates and sulphates) were analyzed by ion chromatography; pH was measured with a pH-meter Cryson-507; electrical conductivity (EC) and total dissolved solids (TDS) were determined using the multi-range equipment Cryson-HI8734 and turbidity was measured with a turbidimeter Dinko-D-110. Samples of 100 ml of each sample of water were filtered using a vacuum system through 0.45 μm membrane filters (Millipore). These filters were placed in Chromocult coliform agar (Merck, Darmstadt, Germany) and incubated at 37°C for 24 h for the *E.coli* growth and at 44.5°C for 24 h to obtain the faecal coliforms. The helminths eggs were measured by Bailenger's method [10].

2.3. Soil analysis

Twelve soil samples from 0 to 20 cm depth were taken in both localizations in December 2005 (before wastewater irrigation) and December 2006 (after treated wastewater irrigation). Soil samples were air-dried for at least 2 days and sieved through a 2-mm nylon mesh before analysis. The organic matter (OM) and total N content were measured by an automatic micro-analyzer; the macroelements, microelements and

heavy metals were determined by ICP after nitric-perchloric acid (2:1) digestion; the anions were analyzed by ion chromatography after aqueous extraction (1:20 w,v); the values of pH were determined in saturated soil pastes and the EC was determined in 1:5 aqueous soil extracts. For soil microbiological analysis, 12 soil samples were taken per localization. Each sample was placed in a sterile recipient with a closing system suitable for isolation of the environment. Samples were transported in an ice chest to the laboratory and stored at 5°C before being processed. The 30 g soil samples were diluted 1:10 in sterile 0.1% peptone water and homogenized by hand in sterile laboratory stomacher bags. The microbiological analyses of these dilutions were done using the techniques described previously for water analyses.

2.4. Leaf analysis

Spring flush leaves from non-fruiting branches were sampled before treated wastewater application (December 2005) and after wastewater irrigation (December 2006). Twenty leaves were sampled from 12 trees at each orchard. Leaves were washed with a special detergent (Alconox 0.1%), rinsed in tap water, cleaned with a dilute solution of 0.005% HCl and finally rinsed in distilled water, left to drain on a filter paper and oven dried for at least 2 days at 65°C. Following digestion in a nitric-perchloric acid, the concentration of macroelement, microelements and heavy metals was determined by ICP, and the concentration of anions was measured by ion chromatography after aqueous extraction. The total N and C concentrations were measured using an automatic microanalyzer.

3. Results and discussion

The analysis of both types of irrigation water showed clear differences in their composition.

Table 1

Physicochemical and microbiological analysis of irrigation water used in both locations (Cartagena and Campotejar). Data contain average values derived from all samples collected during 2006

		Cartagena	Campotejar	<i>t</i> -test	Recommended range
Macroelements (ppm)	Na	334 ± 34	311 ± 15	ns	0–900
	K	42 ± 12	25 ± 10	ns	0–100
	Ca	107 ± 26	69 ± 20	ns	0–200
	Mg	50 ± 10	41 ± 9	ns	0–60
Microelements (ppm)	Fe	0.04 ± 0.02	0.04 ± 0.03	ns	0–1.5
	B	1.39 ± 0.03	0.84 ± 0.03	*	0–1
	Mn	0.03 ± 0.02	0.03 ± 0.01	ns	0–1.5
Heavy metals (ppm)	Ni	0.12 ± 0.03	0.11 ± 0.04	ns	0–2
	Cd	0.03 ± 0.01	0.02 ± 0.01	ns	0–0.05
	Cr	0.02 ± 0.01	0.03 ± 0.01	ns	0–1
	Cu	0.03 ± 0.01	0.02 ± 0.01	ns	0–3
	Pb	0.02 ± 0.01	0.02 ± 0.01	ns	0–1
	Zn	0.11 ± 0.02	0.03 ± 0.02	ns	0–1
Anions (ppm)	Chlorides	221 ± 14	170 ± 12	*	0–100
	Nitrates	3.86 ± 1.36	5.91 ± 1.22	ns	0–50
	Phosphates	3.10 ± 0.60	3.00 ± 0.10	ns	0–15
	Sulphates	354 ± 185	529 ± 167	ns	0–400
Physicochemical parameters	pH	8.28 ± 0.57	7.94 ± 0.71	ns	6.5–8.5
	EC (dS/m)	2.82 ± 0.26	2.10 ± 0.10	*	0.7–3
	TDS (mg/l)	1589 ± 362	945 ± 54	*	450–2000
	Turbidity (NTU)	6.02 ± 1.90	1.89 ± 0.47	*	0–5
Microbiological parameters	Faecal coliforms (UFC/100 ml)	430 ± 125	<10	*	0–200
	<i>E. coli</i> (UFC/100 ml)	<10	<10	ns	0–100
	Helminth (eggs/10 l)	<10	<10	ns	<1

Mean content ($n=12$), *, statistically significant at $P < 0.05$ level of significance.

Cartagena's water showed significantly higher values in EC, TDS and turbidity. This water also had a significantly higher concentration in chlorides and B (Table 1). In general, most of the analyzed elements in Cartagena's irrigation water showed a higher concentration than in Campotejar. This fact results in large part from Campotejar's use of 50% well water mixed with the reclaimed wastewater.

In both locations, the irrigation water was hard with a slightly high pH level. In these conditions, it would be interesting to apply a corrector acid to avoid magnesian and calcic precipitations [11]. These precipitates create drip clogging, that is one of the more important problems associated with the use of reclaimed wastewater in drip irrigation.

Table 2

Physicochemical and microbiological analysis of soils in Cartagena. Data contain average values derived from the samples collected before and after the application of treated wastewater

		Before	After	<i>t</i> -test	Recommended range
Chemical analysis	Organic matter (%)	1.38 ± 0.12	1.40 ± 0.13	ns	0–1.75
	N (%)	0.08 ± 0.02	0.10 ± 0.02	ns	0.13–0.18
	Na (ppm)	1183 ± 183	900 ± 225	ns	0–2000
	K (ppm)	437 ± 44	358 ± 32	ns	190–300
	Ca (ppm)	1460 ± 227	1417 ± 190	ns	0–3000
	Mg (ppm)	487 ± 30	422 ± 48	ns	300–600
	Fe (ppm)	332 ± 86	237 ± 21	ns	100–400
	B (ppm)	206 ± 15	259 ± 12	*	2–200
	Mn (ppm)	177 ± 13	278 ± 15.22	*	250–1700
	Ni (ppm)	0.15 ± 0.03	0.09 ± 0.04	ns	0–10
	Cd (ppm)	0.04 ± 0.01	0.03 ± 0.01	ns	0.01–7
	Cr (ppm)	0.02 ± 0.01	0.21 ± 0.01	*	0–5
	Cu (ppm)	0.76 ± 0.15	1.11 ± 0.01	*	1–10
	Pb (ppm)	3.75 ± 1.34	3.74 ± 1.69	ns	2–200
	Zn (ppm)	3.15 ± 0.49	3.57 ± 0.12	ns	3–20
	Chlorides (ppm)	2063 ± 20	2231 ± 25	*	0–3000
	Nitrates (ppm)	402 ± 32	522 ± 42.20	*	0–1000
	Sulphates (ppm)	839 ± 110	887 ± 167	ns	0–2000
	pH	7.84 ± 0.12	7.93 ± 0.18	ns	6.5–7.5
	EC (dS/m)	0.22 ± 0.01	0.31 ± 0.01	*	0.1–0.7
Microbiological parameters	Faecal coliforms (UFC/100 ml)	<10	7300 ± 250	*	0–1000
	<i>E. coli</i> (UFC/100 ml)	<10	<10	ns	0–100
	Helmints (eggs/10l)	<10	<10	ns	<1

Mean content ($n=12$), *, statistically significant at $P < 0.05$ level of significance.

Salinity problems can appear when the EC of the irrigation water is higher than 1.5 dS/m. Both types of water presented high values of EC, being higher in Cartagena (close to 3 dS/m) than in Campotejar (around 2 dS/m). This salinity problem is especially serious for lemon trees, which are considered to be sensitive crops to salts [12].

The high level of EC observed in our trials was primarily due to the high concentration of chlorides and B in both locations (Table 1). High levels of chlorides in citrus trees can cause a

reduction in vegetative growth and a decrease in the leaf gas exchange [13]. High levels of B can also cause phytotoxic problems in citrus trees [14]. In numerous articles it has been demonstrated that B reduces tree growth and productivity and contributes to defoliation and yellow leaves [15].

The wastewater may also contain significant quantities of toxic metals [16,17] and therefore its long-term use may result in toxic accumulation of heavy metals with unfavourable effects on plant

Table 3

Physicochemical and microbiological analysis of soil in Campotejar. Data contain average values derived from the samples collected before and after the application of treated wastewater

		Before	After	<i>t</i> -test	Recommended range
Chemical analysis	Organic matter (%)	1.59 ± 0.10	1.65 ± 0.23	ns	0–1.75
	N (%)	0.10 ± 0.02	0.12 ± 0.22	ns	0.13–0.18
	Na (ppm)	900 ± 225	683.21 ± 25.22	*	0–2000
	K (ppm)	358 ± 00	380 ± 20	ns	190–300
	Ca (ppm)	1417 ± 190	1510 ± 10.25	ns	0–3000
	Mg (ppm)	422 ± 48	430 ± 4.15	ns	300–600
	Fe (ppm)	237 ± 21	242 ± 25	ns	100–400
	B (ppm)	59 ± 12	134 ± 24	*	2–200
	Mn (ppm)	178 ± 14	171 ± 16	ns	250–1700
	Ni (ppm)	0.09 ± 0.04	0.04 ± 0.02	ns	0–10
	Cd (ppm)	0.03 ± 0.01	0.02 ± 0.01	ns	0.01–7
	Cr (ppm)	0.02 ± 0.01	0.08 ± 0.01	ns	0–5
	Cu (ppm)	1.06 ± 0.21	0.85 ± 0.04	ns	1–10
	Pb (ppm)	3.44 ± 1.69	5.23 ± 1.02	ns	2–200
	Zn (ppm)	3.24 ± 0.12	7.20 ± 0.47	*	3–20
	Chlorides (ppm)	1381 ± 25	1110 ± 26.55	*	0–3000
	Nitrates (ppm)	346 ± 40	210 ± 10.22	ns	0–1000
	Sulphates (ppm)	887 ± 167	1320 ± 141	*	0–2000
	pH	7.93 ± 0.18	7.89 ± 0.18	ns	6.5–7.5
EC (dS/m)	0.09 ± 0.01	0.15 ± 0.03	*	0.1–0.7	
Microbiological parameters	Faecal coliforms (UFC/100ml)	<10	<10	ns	0–1000
	<i>E. coli</i> (UFC/100 ml)	<10	<10	ns	0–100
	Helminths (eggs/10 l)	<10	<10	ns	<1

Mean content ($n=12$), *, statistically significant at $P < 0.05$ level of significance.

growth [18]. In our case, however, the concentration of heavy metals measured in both types of water were always included in the optimum range recommended (Table 1).

Apart from the presence of heavy metals, wastewater is a carrier of bacteria, viruses, protozoa and nematodes, which can cause various diseases, a situation found especially in some developing countries, where they use partially processed wastewater for crop irrigation [19]. In this sense, Campotejar's water microbiological

quality was good because of the absence of microbiological toxicity indicators. However, high levels of faecal coliforms were observed in Cartagena's reclaimed wastewater (Table 1), exceeding the maximum concentration ranges for irrigation recommended by the World Health Organization [7] and the US Environmental Protection Agency [20]. The higher microbiology load in Cartagena's wastewater generated an increase in soil faecal coliforms in this location (Table 2).

Table 4

Leaf mineral analysis in Cartagena. Data contain average values derived from the samples collected before and after the application of treated wastewater

Leaf chemical analysis	Before	After	<i>t</i> -test	Recommended range
C (%)	40.66 ± 0.50	40.78 ± 0.50	ns	
N (%)	2.71 ± 0.07	2.80 ± 0.09	ns	2.4–2.7
Na (ppm)	148 ± 44	123 ± 25	ns	0–3000
K (%)	1.57 ± 0.67	0.90 ± 0.04	ns	0.7–1
Ca (%)	4.53 ± 0.25	5.62 ± 0.76	ns	3–5
Mg (%)	0.31 ± 0.03	0.69 ± 0.04	*	0.25–0.45
Fe (ppm)	144 ± 59	215 ± 23	ns	61–100
B (ppm)	42.90 ± 10.3	54.89 ± 4.64	ns	31–100
Mn (ppm)	26.91 ± 10.28	20.11 ± 2.31	ns	26–60
Ni (ppm)	3.71 ± 0.73	3.26 ± 0.60	ns	1–10
Cd (ppm)	2.22 ± 0.31	1.11 ± 0.87	ns	0.2–3
Cr (ppm)	11.94 ± 0.96	15.50 ± 0.32	ns	0.1–40
Cu (ppm)	12.61 ± 1.65	14.41 ± 2.25	ns	6–14
Pb (ppm)	16.11 ± 0.99	21.01 ± 0.57	*	0.1–40
Zn (ppm)	52.91 ± 14.73	55 ± 1.70	ns	15–200
Chlorides (ppm)	659 ± 120	854.21 ± 123.13	ns	0–1000
Nitrates (ppm)	455 ± 23	442.11 ± 43.44	ns	0–1000
Sulphates (ppm)	864 ± 110	872.42 ± 112.23	ns	0–1000

Mean content ($n=12$), *, statistically significant at $P < 0.05$ level of significance.

The wastewater can constitute a significant plant nutrient source for soils of low fertility [21]. Wastewater may increase K and S levels [20], and may also contribute to the accumulation of organic matter up to 59%. Similarly, it may increase the Fe content of the soil. In our experiment, the reclaimed wastewater did not generate an increase in soil organic matter, macronutrients and Fe (Tables 2 and 3). The pH of the soil samples was found to be within the range of 6.6 to 8.4, which is the most desired range in agricultural soils (Tables 2 and 3).

In the same way, some researchers claim that reclaimed wastewater is an important source of nitrogen for citrus trees [5,22]. In this experiment, it was observed that foliar nitrogen levels were in the optimum range considered for citrus trees development (2.5–2.8%) [23] (Tables 4 and 5).

It is important to emphasize that B and chlorides concentrations founded in Cartagena's soil were significantly higher than Campotejar's soil (Tables 2 and 3). B concentrations exceed the recommended range in Cartagena soil after irrigation with reclaimed water. Mn, Cr and Cu concentrations also increased in Cartagena soil after wastewater irrigation, but the values were maintained in the recommended ranges (Table 2). Na and chloride concentrations decreased with the treated wastewater application in Campotejar soil; however, the soil EC increased due to the significant sulphates concentration increment observed after wastewater application (Table 3).

In spite of high soil B levels and high chloride concentration in both types of water, leaf salt toxicity symptoms were not observed, and the leaf macro-nutrient, micro-nutrient and heavy

Table 5

Leaf mineral analysis in Campotejar. Data contain average values derived from the samples collected before and after the application of treated wastewater

Leaf chemical analysis	Before	After	<i>t</i> -test	Recommended range
C (%)	41.77 ± 0.24	42.02 ± 0.22	ns	
N (%)	2.75 ± 0.31	2.82 ± 0.34	ns	2.4–2.7
Na (ppm)	106 ± 25	123 ± 10.35	ns	0–3000
K (%)	0.86 ± 0.09	0.72 ± 0.02	ns	0.7–1
Ca (%)	4.53 ± 0.37	4.30 ± 0.37	ns	3–5
Mg (%)	0.26 ± 0.02	0.33 ± 0.01	ns	0.25–0.45
Fe (ppm)	111.4 ± 10	123.12 ± 8.57	ns	61–100
B (ppm)	27.10 ± 4.64	30.50 ± 2.12	ns	31–100
Mn (ppm)	30.20 ± 6.57	21.11 ± 3.20	ns	26–60
Ni (ppm)	3.26 ± 0.60	3.11 ± 0.50	ns	1–10
Cd (ppm)	2.31 ± 0.07	2.80 ± 0.23	ns	0.2–3
Cr (ppm)	15.50 ± 0.32	21.20 ± 0.62	*	0.1–40
Cu (ppm)	14.41 ± 2.25	10.11 ± 1.10	ns	6–14
Pb (ppm)	15.21 ± 0.47	17.23 ± 0.90	ns	0.1–40
Zn (ppm)	60.30 ± 1.70	63.45 ± 2.30	ns	15–200
Chlorides (ppm)	669 ± 25	802.11 ± 24.77	*	0–1000
Nitrates (ppm)	401 ± 40	420.12 ± 20.44	ns	0–1000
Sulphates (ppm)	716 ± 167	732.04 ± 52.36	ns	0–1000

Mean content ($n=12$), *, statistically significant at $P < 0.05$ level of significance.

metal concentrations were always in the recommended range (Tables 4 and 5). High leaf accumulation of Mg and Pb was observed after the treated wastewater application in Cartagena, although the Pb concentration was always in the recommended range (Table 4). A leaf accumulation of Cr and chlorides was seen after reclaimed water irrigation in Campotejar, but these leaf mineral accumulations have no toxic effects because they were in the recommended range (Table 5).

4. Conclusions

The mix of reclaimed wastewater and well water used in Campotejar had a better agronomic

and microbiological quality than Cartagena's reclaimed wastewater. Therefore, the possibility to mix reclaimed wastewater with well water is a good solution to avoid the problems associated with wastewater use in agriculture.

The high salinity and B concentration were the main problems associated with treated wastewater used in our experiments. Although leaf toxicity levels were not observed, salt accumulation can be a decisive problem for citrus crops.

In both locations, the treated wastewater application did not increase the macronutrients and organic matter measured in the soil. In our conditions, the wastewater did not constitute a nutrient source for the soil.

Acknowledgment

This paper is funded by the projects IRRIVAL (EC, FP6-FOOD-CT-2006-023120) and RIDECO (Consolider-Ingenio 2010- CSD2006-0067).

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