Comparing the impacts of drip irrigation by freshwater and reclaimed wastewater on the soil microbial community of two citrus species.
Comparación de los impactos del riego localizado con agua del Trasvase o agua regenerada sobre la comunidad microbiana del suelo en dos especies de cítricos.

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Abstract

The search for new water resources for irrigation is a mandatory requirement in Mediterranean agroecosystems. The impacts of irrigation with water from different origins were evaluated in the soil microbial community and plant physiology of grapefruit and mandarin trees in the south-east of Spain. The following treatments were considered: i) freshwater from the Tajo-Segura water transfer canal and well (TW) ii) reclaimed water from a wastewater treatment plant (RW) iii) irrigation with TW, except in the second stage of fruit development when RW_c was applied iv) irrigation with RW, except in the second stage of fruit development when TW_c was applied. Enzyme activities and phospholipids fatty acids were determined to know the phylum of microbial biomass.

Keywords: Enzyme activities; semiarid, plant physiology.

Resumen

La búsqueda de nuevos recursos para el riego es un requerimiento en los agro ecosistemas mediterráneos. Se evaluó el impacto del riego con aguas de distintos suministros sobre la comunidad microbiana del suelo y sobre la fisiología de la planta en cultivos de pomelo y mandarino en el Sureste de España. Se consideraron los siguientes tratamientos: i) Agua procedente del Trasvase Tajo-Segura y de pozo (TW) ii) Agua regenerada procedente de una EDAR de la zona (RW) iii) Riego con TW, excepto en la segunda fase de desarrollo del fruto en la que se regó con RW_c iv) Riego con RW, excepto en la segunda fase de desarrollo del fruto en la que se regó con TW_c.

Palabras clave: Actividades enzimáticas; semiárido; fisiología de planta.

1. INTRODUCCIÓN

In Mediterranean regions, water availability is predicted to decrease in the coming decades. The region of Murcia, located in the South-east of Spain, is characterized by a deficit of water resources that reaches 400 Hm^3 in the hydrographic basin (PHCS, 2016). In these conditions farmers needs to use non-conventional water resources for irrigation and reclaimed water from a wastewater plant is a feasible option for agriculture. This water has the problem of containing an excess of salt that may increase the electrical conductivity and the risk of soil salinization [1] and conversely has available sources of organic matter that could improve the productivity in agricultural areas. Hence, irrigation with combined wastewater and freshwater can be an adequate solution to the problems commonly related to wastewater use in Citrus sp. agroecosystems, such high salinity and boron concentration.

In the other hand, soil is the fundamental substrate of agriculture. Within soil, microorganisms are greatly responsible of the dynamics of organic matter which remain fundamental to crop yield and soil sustainability [1]. Moreover soil microbial properties can act as early-warning indicators of changes in ecosystems [2].

In this sense, the potential negative effects of wastewaters in plants can be early detected by soil microbial parameters.

Traditionally, the activity of microorganisms has been evaluated by microbial-ecosystem indicators such as soil respirations and enzyme activities involved in the cycles of C, N and P [3, 4].

For a water management perspective, this study aims to answer the following question: is it better the irrigation with wastewater alone or in combination with fresh water? In this respect, combinations of water from different sources might represents a proper

2. MATERIALES Y MÉTODOS

2.1 Experimental area, irrigation treatments and soil sampling

The experiment was carried out in Campotéjar-Murcia, Spain with a Mediterranean semiarid climate. The annual reference evapotranspiration (ET_0) and rainfall are, on average, 1326 and 300 mm, respectively. Within this area an orchard of 1 Ha was cultivated with 2 crops. One crops consisted of a 16 year old mandarin trees (*Citrus clementina cv. Orogrande*) grafted on Carrizo citrange (*Citrus sinensis* [L.] osb x *PoncirusTrifoliata* [L.]) rootstock. The other crop consisted of 11 year old "*Star Ruby*" grapefruit trees grafted on *Macrophylla* rootstock (*Citrus macrophylla*). Four irrigation treatments were established:

i) freshwater from the Tajo-Segura water transfer canal and well (TW)
ii) reclaimed water from a wastewater treatment plant (RW)
iii) irrigation with TW, except in the second stage of fruit development when (RWc) was applied
iv) Irrigation with RW, except in the second stage of fruit development when (TWc) was applied.

The trees were irrigated at 100% ET_c from January to December. The replicated plots (n=3) for each treatments and crop were established. A composite soil sample under the canopy of one tree for each sample was composed of six subsamples.

2.2 Water characterization and sampling

The soil within the first 90 cm depth had a loamy texture (24% clay, 33% loam, and 43% sand), with an average bulk density of 1.37 g cm^{-3}. Before the experiment, the soil electrical conductivity (EC) was 2.1 dS m^{-1}. Samples were taken to a depth of 20 cm in October 2015, before harvesting the fruit. The EC_w was 1 ds/m for TW and 3 21 dSm^{-1} for RW. SAR (meq/l) was 1,39 (TW) and 9,45 (RW)

2.3 Plant water status and gas exchange parameters.

The stem water potential was analyzed monthly at midday using a pressure chamber (model3000; Soil Moisture Equipment Corp., Santa Barbara, USA). Two mature leaves from the canopy were selected. At least 2 h before the measurement, the leaves were covered with aluminum foil and enclosed within polyethylene bags.

2.4. Data analysis

The normality and homogeneity of the variables were checked by the Kolmogorov-Smirnov and Levene tests, respectively. The variables were transformed logarithmically when necessary. Data were analyzed using two-way ANOVA with irrigation treatment and crop as main factors. Differences were considered significant at $P < 0.05$.

3. RESULTS AND DISCUSSION.

Significant differences between TW and RW water were observed: RW had greater salinity and sodicity, with average values of EC_w around 3.21 dS m-1 and SAR_w around 9.45 meq/l, whereas TW had lower values of EC_w, 1.00 dSm^{-1} and SARw 1.39 meq/ l. The RW water also had higher concentrations of NO_3^-, PO_4^{3-}, SO_4^{2-}, Cl^-, K, B, and Na than TW. The concentrations of organic C and N in the reclaimed water were 18 and 7 mg/l, respectively.

Overall, the bacterial and fungal soil PLFA contents were higher under grapefruit than under mandarin trees. In the case of grapefruit, TW produced a lower bacterial PLFA content than RW, RWc, or TWc. In contrast, RW showed the lowest values in the mandarin trees (Fig. 1).

The treatment and crop, and their interaction, influenced significantly the net photosynthesis (A) and stomatal conductance (gs). The stem water potential (Ψs) was affected significantly by crop. Lower annual average values of A and gs were observed in plants irrigated with RW, RWc or TWc, in comparison to TW (Table 2).

With the exception of RW, soil respiration was higher in grapefruit soil samples than in mandarin ones. Respiration was greatest in TWc soil in the case of grapefruit, and in RW in the case of mandarin (Table 3). The enzyme activities were higher in grapefruit soil than in mandarin soil, with the exception of β-glucosidase, cellobiohydrolase, and p-phenol oxidase for TWc.

4. CONCLUSION

Irrigation with reclaimed water did not negatively impact the soil microbial community of semiarid soils under grapefruit and mandarin crops. Indeed, the annual use of reclaimed water or the dual irrigation with TWc influenced positively the microbial biomass and biogeochemical activities of soil microbial communities under grapefruit. The mandarin community seemed more sensitive to the annual irrigation with RW but, overall, responded positively to dual irrigation. Changes in biomass and activity were coupled to variations in the structure of the microbial community. These microbial responses were probably shaped by the specific plant physiology, the rootstock sensitivity to salinity and water relations of the crop.

5. ACKNOWLEDGEMENTS

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Table 1. Soil chemical parameters, basal respiration, and microbial ratios in grapefruit and mandarin irrigation treatments.

	Grapefruit				Mandarin			
Parameter	**TW**	**RW**	**TW_c**	**RW_c**	**TW**	**RW**	**TW_c**	**RW_c**
TOC	0.99±0.11a*	0.92±0.01a	1.10±0.06b	0.91±0.07a*	0.75±0.04a	0.81±0.07a	1.24±0.07b	0.77±0.09a
Total N	0.19±0.01b*	0.14±0.01a	0.19±0.01b*	0.12±0.02a*	0.13±0.01a	0.14±0.01a	0.26±0.05b	0.16±0.01a
EC	616±187a	2486±221d	1786±98b	2070±143c	839±91a	2316±173d	1326±84b	1770±95c
pH	8.01±0.24a	8.43±0.27a	8.11±0.15a	8.09±0.19a	8.18±0.22a	8.30±0.17a	8.30±0.24a	8.41±0.20a
Respiration	25.21±1.39a*	29.98±2.14b	47.08±4.98c*	28.09±2.01b*	13.88±0.87a	31.40±2.54c	20.85±3.27b	13.86±0.55a
Resp/TOC	15.21±1.19a*	19.77±2.21a*	23.52±0.97b*	18.98±2.73a*	12.59±2.35b	25.53±2.47c	8.55±1.48a	11.73±1.14b
B/F	2.96±0.26a*	3.44±0.13b	3.43±0.17b	3.26b±0.21b*	3.50±0.14a	3.76±0.16b	3.35±0.01a	3.70±0.20b
G+/G-	0.93±0.078ab*	0.86±0.11a	1.15±0.094b*	0.83±0.10a*	1.21±0.10b	0.77±0.12a	1.04±0.097b	1.13±0.083b

Table 2. The physiology, water status and yield of evaluated crops

	Grapefruit				Mandarin			
Parameter	**TW**	**RW**	**TW_c**	**RW_c**	**TW**	**RW**	**TW_c**	**RW_c**
A	12.91±0.33bc*	12.31±0.37c*	11.55±0.89b*	9.76±0.94a*	8.77±0.29b	6.81±0.32a	8.31±0.32b	6.95±0.56a
g_s	0.16±0.01c*	0.13±0.01bc*	0.12±0.01b*	0.10±0.01a*	0.086±0.01b	0.062±0.01a	0.070±0.01ab	0.065±0.01a
Ψ_s	1.083±0.84a*	1.13±0.94a*	1.11±0.97a*	1.13±0.94a*	0.80±0.37a	0.81±0.38a	0.81±0.47a	0.85±0.45a
Y	96.67±12.25a*	101.67±14.72a*	91.25±11.58a*	103.33±15.97a*	35.86 ± 3.47a	31.21 ± 2.14a	45.26 ± 3.51b	36.54 ± 2.87a

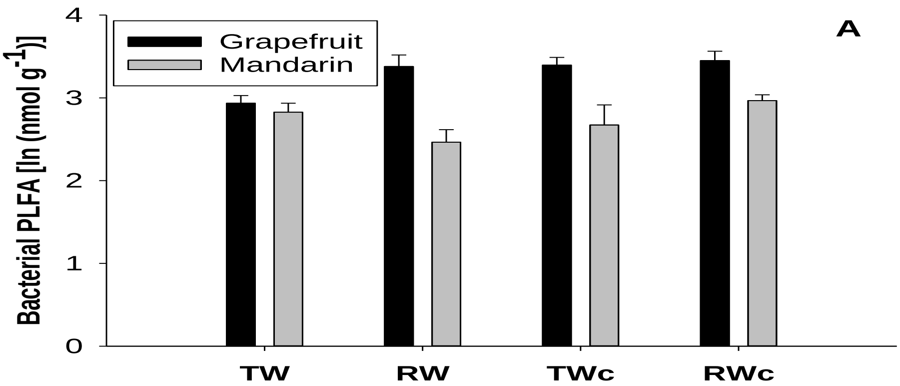

Figure 1. Bacterial and fungal soil PLFA contents.