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Combined effects of reduced irrigation and water quality on the soil microbial community of a citrus orchard under semi-arid conditions

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ABSTRACT

Sustainable agriculture in Mediterranean areas is compromised by the structural deficit of water resources. Under this situation, the impacts of alternative water managements on the microbial community, as a critical component of the soil quality, need to be properly understood. We evaluated the long-term impacts of irrigation systems differing on the quantity and quality of water, and their interactions, on the biomass (phospholipid fatty acid analysis), diversity and composition (16S rRNA gene profiling), and enzyme activities of the soil microbial community of an orchard cultivated with grapefruit trees in South-East Spain. The impact of water quantity was evaluated by irrigation with optimal amount of water or by irrigation with a reduced volume of water in the temporal frame when the crop is less sensitive, so-called regulated deficit irrigation (RDI). The impact of water quality was evaluated attending to the source of the irrigation water: water from a river channel-transfer (TW) or reclaimed water from a wastewater treatment plant (RW). Electrical conductivity was higher in soils irrigated with RW than in soils irrigated with TW. The content of total organic C in the soil was affected by water quality but not by water quantity. Soils irrigated with TW showed higher total organic C than soils irrigated with RW. As in the case of plant productivity, RDI had a negative impact on plant productivity, soil microbial biomass and enzyme activities in summer. This finding indicates a slow-down of organic matter decomposition under restricted irrigation. Bacterial biomass was more sensitive to RDI when RW was used, whereas the fungal biomass was more sensitive to RDI when TW was used.

Bacterial diversity and plant productivity were more sensitive to water quantity than to water quality. The increase of the abundance of *Proteobacteria* and *Bacteroidetes* in soils irrigated with RW in summer suggested a higher resilience of this treatment mediated by copiotrophic organisms. A recovery of the enzyme activity and microbial biomass of soils irrigated with RW and RDI was observed in January and June. The resilience of biogeochemical and the microbial biomass processes after RDI coursed through changes in the structure of the microbial community as revealed by the multivariate analyses of fatty acids. The utilisation of reclaimed water during RDI promoted a more-resilient community that translated into a recovery of microbial biomass and enzyme activities after the water restriction ended. These results imply potential ecological benefits of the irrigation with reclaimed water that should be considered under the water limitation predicted in climate change models in Mediterranean areas.

1. Introduction

The south-east of Spain is characterised by a structural deficit of available water resources that reaches 606 Mm^3 (Ibor et al., 2011). Hence, it is clear that water scarcity is a limiting factor for the development of sustainable agriculture in Mediterranean areas. Indeed, the problem will be magnified in the coming decades as a consequence of climate change, due to a tremendous decrease in precipitation and a rise in temperatures and evapotranspiration (IPCC, 2013).

Under these conditions, the water resources will not be enough to satisfy sustainable agriculture in semi-arid agro-ecosystems.

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Therefore, alternative systems of water management need to be proposed. Farmers are seldom subjected to a dichotomy: whether handling a water deficit or utilising non-conventional water resources (desalinated or reclaimed water) for irrigation. In the first case, one option could be the reduction of the volume of water used for irrigation in the temporal frame when the crop plant is less sensitive, the so-called "regulated deficit irrigation" (RDI). In the second case, the use of reclaimed waters can provide a continuous and cheap source of water for agriculture (Mounzer et al., 2013; Pedrero et al., 2014). The impacts of these alternative water management systems have been studied at the levels of plant physiology and productivity (Mounzer et al., 2013; Pedrero et al., 2014, 2015; Nicolás et al., 2016). However, much less information is available about the impacts of water management on the soil microbial community. This lack of knowledge constitutes a critical issue because the microbial community is a key attribute for soil quality and microbes carry out fundamental processes related to soil fertility (Bastida et al., 2008a; Zornoza et al., 2015). For instance, they are responsible for the cycling of organic matter and the generation of nutrients for plants through their enzymatic machinery (Nannipieri et al., 1990). It is clear that accumulated damage to soil would impact plant productivity irreversibly.

The impacts of irrigation with reclaimed waters on the soil microbial community have been partially evaluated and show contrasting results (reviewed by Becerra-Castro et al., 2015). On the one hand, reclaimed waters may contain contaminants and often have a high salt concentration that may increase the risk of soil salinisation, with the consequent decrease of microbial biomass and enzyme activities (García and Hernández, 1996). On the other hand, reclaimed waters often contain soluble organic matter that may benefit the development and enzyme activities of the soil microbial community (Gelsomino et al., 2006; Adrover et al., 2012; Elifantz et al., 2011), with potential changes in its composition (Frenk et al., 2014; Wafula et al., 2015).

It is known that water availability plays a role in the performance of soil microbial communities in natural and agricultural ecosystems at the level of microbial growth and biomass (Fierer et al., 2003; Hueso et al., 2011, 2012; Meisner et al., 2013); microbial composition (Hawkes et al., 2011; Placella et al., 2012) and biogeochemical cycles (Placella et al., 2012; Goransson et al., 2012). Indeed, several studies have highlighted that drying-rewetting dynamics impact strongly the decomposition of organic matter in agroecosystems (Birch, 1958, 1964; Austin et al., 2004; Rey et al., 2005). However, from an agriculture management perspective, the impacts of regulated deficit irrigation on the soil microbial communities have been less studied. An integrative study considering also plant productivity would reinforce the relationships between plants and soil microbes in Mediterranean agroecosystems.

Here, we aim to comprehensively understand the impacts of irrigation water quantity (RDI or optimal water amount) and quality (transferred water of high quality or reclaimed water) on the biomass, enzyme activity and composition of soil microbial communities in a Mediterranean grapefruit orchard. These responses will be linked to plant productivity. Furthermore, we will evaluate the legacy effects of RDI and the resilience of the soil microbial community in terms of biomass generation and biogeochemical potential. We seek to respond to the following pragmatical question: "which is better: a reduced amount of high-quality water or an optimal amount of reclaimed water?" For this purpose, we will monitor extracellular enzyme activities related to the biogeochemical cycles of elements (Bastida et al., 2008a; García and Hernández, 1996) and phospholipid-fatty acids (PLFAs), as indicative of the biomass of the soil microbial community (Frostegard et al., 1993; Bastida et al., 2008b, 2015; Nicolás et al., 2014), and use 16S rRNA gene profiling of the microbial communities to understand the impacts of water management on the composition, structure and diversity of the soil microbial community (Fierer et al., 2007; Bastida et al., 2013).

We hypothesised that the salinity of reclaimed water would impact negatively the biomass of microbial communities, but also that this effect might be counteracted by its content of available organic matter. Decreasing the amount of water used for irrigation (RDI) in summer will decrease the biomass and activity of the microbial communities in this season and will promote the development of specific microbial populations, depending on the quality of the water. Moreover, we expect that the resilience of the soil microbial community after the episode of restricted irrigation in summertime will depend on the quality of the water used for irrigation.

2. Material and methods

2.1. Experimental design and irrigation treatments

The experiment was developed in a commercial 0.5-ha orchard cultivated with 11-year-old 'Star Ruby' grapefruit trees (*Citrus paradisi* Macf) grafted on Macrophylla rootstock [*Citrus macrophylla*]. This orchard is located in Campotéjar-Murcia, Spain (38°07′18″N; 1°13′15″W) and is characterised by a Mediterranean semi-arid climate with warm, dry summers and mild winter conditions. The average annual reference evapotranspiration (ET_0) and rainfall were 1326 and 300 mm, respectively. 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 1.41 g cm^{-3}.

The experiment took the form of three completely-randomised plots per irrigation treatment (for a total of 12 plots). Each plot was made up of 12 trees (288 m^2), organised in three adjacent rows with four trees per row. In each plot, the two central trees of the middle row were used for evaluation of plant production.

The agronomic design of the irrigation treatments is described elsewhere (Pedrero et al., 2015). Four irrigation treatments, based on the water quality of the irrigation source and water quantity, were performed. One source (TW), with an average electrical conductivity of 1.1 dS m^{-1}, was pumped from the "Tagus-Segura" water transfer canal, which supplies the major part of the water used for irrigation in south-east Spain. The other source was tertiary reclaimed water (RW) pumped from a nearby wastewater treatment plant. This source was automatically blended, at the irrigation control-head, with water from the canal to maintain a constant electrical conductivity of around ~3 dS m^{-1} throughout the experiment.

From 2005 to 2007 the whole orchard was fully irrigated with water transferred from the channel (TW). The following irrigation scheme was performed from 2008 onwards. The control treatments involved irrigation with TW or RW during the whole season at 100% ETc (TW-C and RW-C, respectively). The RDI treatment consisted of irrigation at 100% ETc, except during the second stage of fruit growth, for 55–65 days between late-June and mid-September, when it consisted of 50% of the amount of water applied to the control (TW-RDI and RW-RDI). It means that TW-RDI and RW-RDI soils sampled in August received 50% of the water utilized in TW-C and RW-C during approximately two months of each of the last 7 years. The total amounts of water applied were measured with inline water flow meters and in the control treatments were 614 (TW-C) and 593 (RW-C) mm, whilst in the RDI treatments were 518 (TW-RDI) and 493 (RW-RDI) mm, respectively, Hence, RDI treatments meant reductions of about 16% of water per year.

All treatments included application of the same amounts of fertiliser ($N-P_2O_5-K_2O$), applied through the drip irrigation