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Treated agro-industrial wastewater irrigation of tomato crop: Effects on qualitative/quantitative characteristics of production and microbiological properties of the soil

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ABSTRACT

A comparative study was carried out to evaluate the effects of two water irrigation sources on the quality and microbiological safety of tomato plants and fruit, and on the microbiological soil properties: irrigation with groundwater (GW) and with treated agro-industrial wastewater (TW). In a field experiment in southern Italy (Apulia region), the physico-chemical characteristics of the irrigation waters and the fruit quality parameters were determined. *Escherichia coli*, fecal *Enterococci* and *Salmonella* spp. were also monitored in the irrigation waters, tomato plant and fruit, and root-zone soil. Bacteriological analysis for total heterotrophic counts (THCs) were determined for plant, fruit, and soil samples. The irrigation water source did not significantly affect yield quantitative traits. However, with GW, the marketable fruit yield was higher than with TW (~82 vs. ~79 Mg ha^{-1}, respectively). For both irrigation treatments, the most important qualitative parameters that characterize the processing tomato fruit (i.e., dry matter content, pH, soluble solid content, color parameters) were in agreement with reports in the literature. For the microbiological results, the mean levels of *E. coli* and fecal *Enterococci* were 4408 and 3804 CFU 100 ml^{-1}, respectively, for TW (above the Italian guidelines for TW re-use). For the tomato plant and fruit, no *E. coli* isolated in either, and fecal coliforms and THC were not influenced by the irrigation waters ($P > 0.05$). Total bacterial enumeration by quantitative PCR was lower in soil irrigated with GW, than TW (3.69 vs. 4.02, $\times 10^6$, respectively). Moreover, soil microbial community patterns substantially differed between the two water treatments. These data show that while fecal indicators are not affected, the community composition and dynamics of the whole bacterial population in soil is influenced by the different qualities of these waters used for irrigation.

1. Introduction

The re-use of wastewater in agriculture is gaining wider acceptance in many parts of the world. It represents an agronomic option that is increasingly being investigated and taken up in regions with water scarcity, growing urban populations, and rising demand for irrigation water (Meli et al., 2002; FAO, 2011). Many irrigated areas around the world are experiencing water shortages due to several factors, such as climate change and surface and groundwater pollution. Water scarcity poses serious economic, social and even political concerns in all of its aspects. Under these circumstances, treated wastewater use can help to mitigate the damaging effects of local water deficits (FAO, 2010).

Treated wastewater not only offers an alternative water irrigation source, but also the opportunity to recycle plant nutrients (Chen et al., 2008). Its application might ensure the transfer of fertilizing elements, such as nitrogen (N), phosphorous (P), potassium (K^+), organic matter, and meso-nutrients and micro-nutrients, into agricultural soil (WCED, 1987). Hence, wastewater nutrients can contribute to crop growth, although there is a need for their periodic monitoring, to avoid any imbalance in the nutrient supplies, which might cause excessive vegetative growth, uneven plant and/or fruit maturity, and/or reduced qualitative/quantitative aspects of yields (Pedrero et al., 2010).

Treated wastewater can also be a source of pathogenic organisms and potentially hazardous chemical substances, such as enteric bacteria and viruses, salts, heavy metals, and surfactants.

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These might then accumulate in the soils, with unfavorable effects on crop quality and productivity, and on the ecological soil conditions (Siebe and Cifuentes, 1995; Chen et al., 2008). One of the major concerns with wastewater re-use is the risk of the transfer of pathogenic microorganisms that represent a potential risk to human health if they enter the food chain (Al-Lahham et al., 2003; Salgot et al., 2006; Toze, 2006; Palese et al., 2009). Indeed, many studies have shown that microbiological contamination can be a major issue for the re-use of treated agricultural wastewater (Rubino and Lonigro, 2008; Lopez et al., 2010; Patterson et al., 2011; Vivaldi et al., 2013). To maximize the benefits and at the same time, to minimize the risks related to the use of treated wastewater, international policies and uniform legislative frameworks should be adopted.

In Italy, the agricultural use of reclaimed wastewater (municipal and agro-industrial) is regulated by Ministerial Decree no. 185/2003. With regard to microbiological contamination levels in particular, this Decree has defined some significantly lower threshold values (e.g., *Escherichia coli*, <10 CFU 100 ml^{-1} in 80% of the samples) than those included in international guidelines. These threshold values can be considered highly restrictive, because the risk of contamination has been reported to be low when contamination of irrigation water does not exceed 1000 CFU ml^{-1} (WHO, 2006; Blumenthal et al., 2000).

Studies have been carried out in southern Italy relating to treated urban wastewater re-use for the irrigation of crops (Pollice et al., 2004; Lonigro et al., 2007; Lopez et al., 2007). These have included wastewater with microbiological contamination levels higher than those required by the current legislation, and they have indicated the opportunity to increase the threshold values in the Italian guidelines. Therefore, there is the need for further studies to better define acceptable microbiological contamination levels of different sources of irrigation water as used on different crops. These should also take into account wastewater treatment, irrigation methods, and cultivation practices.

The majority of the studies conducted on wastewater applications in agriculture have focused mainly on reclaimed urban effluents. The aim of the present study was to determine the effects of secondary treated agro-industrial wastewater on tomato crop performance. In particular, the objectives of the study were: (i) to evaluate the effects of the wastewaters on qualitative and quantitative aspects of tomato crop production; (ii) to assess the impact of the wastewaters on the microbiological contamination of tomato fruit and the microbiological soil properties.

2. Materials and methods

2.1. Field characteristics and agronomic conditions

This field trial was carried out with the tomato (*Solanum lycopersicum* L.; formerly *Lycopersicon esculentum* Mill.) cultivar 'Manyla' (Semillas Fitò, Spain) during the growing season of 2012 (April to August). It took place in an agricultural area in the Foggia district (Stornarella: 41° 15′N, 15° 44′E; altitude, 154 m a.s.l.) of the Apulian region in southern Italy, on a site belonging to the Fiordelisi agricultural and food manufacturing company, which produces and processes vegetables. The tomato plants were grown under a net house structure, covered with an anti-hail net, in six identical 15 m × 30 m plots that were located near to the company wastewater treatment plant.

The experimental area is characterized by a Mediterranean climate, with long-term mean annual rainfall of 590 mm, which is mainly distributed from October to April (Caliandro et al., 2005). The mean monthly main climate parameters recorded during the trial are reported in Table 1. These were measured by a weather station near to the experimental area, and stored on a nearby data-logger (Campbell Scientific, USA). The mean maximum and mean minimum temperatures during the growing season were 34.5 °C and 8.5 °C, respectively, and the total rainfall was 108.4 mm, of which about 62% (67.0 mm) occurred in the first month of the growing season.

The trial was carried out in a clay loam soil (United States Department of Agriculture classification), with a field capacity (−0.03 MPa) of 30.5% dry weight (dw), a wilting point (−1.5 MPa) of 15.9% dw, and a bulk density of 1.41 Mg m^{-3}. The main characteristics of the soil layer of the experimental site (0–60 cm) are as follows: sand, 40.1%; loam 32.5%; clay 27.4%; organic matter 1.6%; Olsen P_2O_5, 80.1 mg kg^{-1}; Ac-extractable K_2O, 730 mg kg^{-1}; total N, 0.8‰ (Kjeldahl); mineral NO_3–N, 4.75 mg kg^{-1}; mineral NH_4–N, 7.50 mg kg^{-1}; pH 7.9; electrical conductivity, 0.49 dS m^{-1}.

The tomato seedlings were transplanted into the plots on April 12, 2012, in mulched paired rows (40 cm apart) spaced at 250 cm, with the plants at a distance of 30 cm apart along each single row. The final plant density was 2.7 plants m^{-2}. The plants were grown in a vertical setting, using nylon threads disposed between plants collar and iron wires arranged longitudinally in the direction of the plant rows, and fixed to the upper part of the nethouse, at 2.5 m from the ground.

During the cropping season, standard agronomic practices for tomato crops in the area were performed. The soil was subsoiling to a depth of 45 cm, and before transplanting, its surface was milled. Pre-transplanting fertilization was applied to the soil by distributing 35 kg ha^{-1} N and 70 kg ha^{-1} P_2O_5. Throughout the crop cycle, 75 kg ha^{-1} N and 100 kg ha^{-1} P_2O_5 were added through fertirrigation. Pest and weed control were performed according to local management practices.

The tomato fruit were hand harvested at full stage maturity. Four harvestings were performed from June to August, on the days after transplanting of: 82 (HD_1), 96 (HD_2), 110 (HD_3) and 124 (HD_4).

2.2. Treatments and experimental design

Two experimental irrigation treatments were applied to the tomato plants: irrigation with groundwater (GW), and irrigation with treated agro-industrial wastewater (TW). The GW was from a water source that is commonly applied for crop irrigation in the experimental area. The TW used in this study was taken from the wastewater treatment plant that purifies all of the wastewater produced by the company during their industrial processing of vegetables (i.e., tomatoes, egg plants, courgettes, peppers). It is an activated sludge wastewater treatment plant that produces an annual volume of effluent of approximately of 46,500 m^3. The incoming wastewaters undergo a preliminary treatment through a 6-mm sieve screen, to separate out the coarse organic waste. The effluent water then goes into an equalization tank, for the secondary biological treatment. At the end of this phase, the wastewater is clarified in a secondary settler, and the sludge is separated out. For the present study, part of the treated wastewater not subjected to chlorine treatment was directed into the experimental area through a 100-mm diameter PVC pipe, and stored in a 3000-l tank; subsequently, it was used for the tomato irrigation.

The experiment was laid out in a randomized complete block design with the two irrigation treatments each replicated three times (Fig. 1). A drip irrigation system was used for the crop irrigation. This comprised a single pipe, with drippers at a 2 l h^{-1} flow rate spaced every 40 cm, and it was arranged in the middle of each paired row. Except for the first irrigation that was designed for the rooting and establishment of the plants, the following irrigations were performed with each water treatment every time the available soil moisture was depleted to the threshold value of 40% (Allen et al., 1998). This irrigation scheduling took into account continuous measurements of volumetric soil water content changes at