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Effect of Irrigation with Treated Wastewater on Potatoes' Yields, Soil Chemical, Physical and Microbial properties

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Abstract

In this study, the effects of irrigation with treated wastewater (TWW) on the physical, chemical, microbiological, and yield of potatoes were investigated. Potatoes (*Solanum tuberosum*) were irrigated by drip irrigation system using conventional irrigation water (CIW), TWW, and blended irrigation water (BIW). The concentration of all chemical and microbial characteristics of irrigation water was falling within the limits of Jordanian standards (JS893/2021), except for turbidity and boron. Pathogen indicators, *Salmonella*, and *Helminth* eggs were not found in TWW. The TWW-irrigated plots were significantly higher than BIW and CIW in electrical conductivity, organic carbon, total nitrogen, and sodium adsorption ratio (SAR). Soil iron, total *Coliform*, and *Escherichia coli* (*E,coli*) contents increased significantly within TWW-irrigated plots. Potatoes' fresh yield weight irrigated with TWW was significantly higher compared to the CIW. Treated wastewater and BIW treatments tended to have more considerable fruit weight and size than CIW. The *E. coli* was not significantly different on the surface of potato fruits, while total *Coliform* increased significantly for fruits within the TWW-irrigated plots.

Keywords: *Treated wastewater, Blended water, Escherichia coli, Pathogens, Salinization, Potatoes, Jordan.*

1. Introduction

Jordan has one of the lowest water availability rates in the world. Since 1964, the Jordanian individual's share of annual water use has decreased from 3,600 to less than 100 m^3/capita, which is less than 10% of the estimated worldwide water poverty level of 1,000 m^3/capita (MWI, 2019). Jordan's Ministry of Water and Irrigation adopted National Strategic Plan (2016-2025) that incorporates blended irrigation water (BIW) with treated wastewater (TWW) a water source added to the water budget for unrestricted reuse utilization for agricultural irrigation. Agriculture uses 52% of the total conventional water in the country; 29% of the irrigation water comes from TWW (MWI, 2020).

Using TWW for agriculture irrigation could be associated with potential problems such as health hazards, salinity build-up, and toxicity hazards (Hashem and Qi, 2021; Qiu et al., 2015). To safeguard public health and make TWW use in agriculture safe, the Jordan Standards and Metrology Organization (JSMO) developed and issued the Jordanian Standards 893/2021 (JSMO, 2021), based on WHO guidelines (WHO, 2006). These standards prohibit the use of TWW for irrigating vegetables that are eaten raw (uncooked).

Although wastewater contains nutrients important for soil productivity, it may contain toxic materials that may influence soil health and crop yield, besides pathogens (Ahmad et al., 2016). Several studies evaluated the positive and negative impact of TWW-irrigation on soil quality and yield, (Chaganti et al., 2021; Jahany and Rezapour, 2020; Paudel et al., 2018; Urbano et al., 2017; Akhtar et al., 2012; Gharaibeh et al., 2016; Cirelli et al, 2012). Furthermore, some studies show that there is a possibility for the transport of pathogens through the leaf, stem, cracks, or flaws in the skin (Serrano et al., 2014; Tournas, 2005). Soil salinization related to irrigation with wastewater is still a concern (Bedbabis et al., 2014). In arid and semiarid areas, salinization is a common problem. Around the world, 0.3–1.5 million hectares of arable land are abandoned each year as a result of salinization (Harper et al., 2021). Therefore, scientific researchers suggested measures to mitigate problems related to TWW irrigation (Nogueira et al., 2013; Cirelli et al, 2012)

The entire world cultivates and consumes potatoes and their production exceeded 370 million tons per year (FAO, 2019). In Jordan, potatoes were the second area planted vegetable after tomatoes in 2019 with a total production of 379 thousand tons (Suleiman, 2022). Potatoes production is concentrated in Jordan Valley, where the irrigation water is from King Talal Dam (KTD). Some potential sources of contaminants affecting the water quality of KTD, contaminants include discharges from the Samra wastewater treatment plant (WWTP) and Wadi Rmemeen (Al-Taani et al., 2018). Although the fact that potatoes are eaten cooked, and subsequently safe, high microbiological irrigation water quality is required because the high and constant humidity beneath the potato crop canopy is conducive to pathogen growth (Adams and Stevenson, 1990). Jordan standard (893/2021) for TWW use for irrigation includes root and tuber crops under the same category as other vegetables that are eaten cooked.

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This study aimed to investigate the effects of TWW used for agriculture irrigation on soil chemical, physical, and microbial properties, and potatoes' yield.

2. Materials and Methods

2.1. Study area and experimental conditions

The experiment was conducted on a farm located in Deir Alla (32.233615°N, 35.603982°E) at an elevation of -224 m below sea level. Summers in Deir-Alla are hot and dry, and winters are mild and wet (Kool, 2016), with an annual mean temperature of 23.6°C. The temperature in summer is around 40°C and rarely drops below 20°C in winter. The average total annual rainfall is 285 mm (Kool, 2016; Tarawneh and Kadıo˘glu, 2003).

A randomized complete block design (RCBD) with three replicates, was used to examine treatments: (i) CIW; (ii) BIW (50% CIW blended with 50% TWW); (iii) TWW. Three separate tanks and three irrigation pumps were used. Each block was distributed in a random order so that they would not be next to each other. Potato tubers (*Solanum tuberosum*) cv. (Florice) was planted in clay loam soil, in January 2021. Every row was irrigated using drip irrigation covered with plastic mulch.

Temporary plastic tunnels were used, at the time of rainfall, to prevent rainwater from entering the treatment plots. Each plot contained three rows, each 7 m long. The plants were spaced 40 cm apart and rows were separated by 130 cm. Each row had an irrigation line, and 6 liters hr^{-1} discharge emitters. Potatoes were harvested in April 2021. TWW was sourced from the secondary stage of the Kufranjah-wastewater treatment plant (KWWTP). The KWWTP plant comprises preliminary treatment (screening, grit removal), activated sludge, tertiary treatment, and sludge treatment.

In this study, the fertilizer requirement for potatoes was divided into three applications throughout the growing season. In the first growth stage, nitrogen-phosphorus-potassium granular (NPK 18-18-5) was applied at the planting date. In the second stage, phosphorus is required at higher levels, so fertilizer (NPK, 12-61-0) is applied while supplying low nitrogen levels to sustain vegetative growth. In the maturity stage, NPK 13-0-46 was applied. The fungicide (Ultimatrix 52.5%WG) was used to control the early blight of potatoes.

For each irrigation event, the irrigation depth was applied and recorded. Farmer's experience was the decisive factor in the timing and amount of irrigation to meet crop water requirements.

2.2. Water analyses

During the experiment (January, February, and March 2021), water samples were collected from the holding tank in clean plastic bottles. Water sampling was conducted whenever the tanks are filled. According to the American Public Health Organization's standards (APHA, 2005), electrical conductivity (EC) was immediately measured using a conductivity meter (Jenway Conductivity Meters); pH using a pH-meter (Mettler Toledo, model FP20 Meter); dissolved oxygen (DO) was measured using a dissolved oxygen meter (Lovibond SD 400 Optical); A turbidity (TUR) meter (Mettler Toledo FSC402) was used to measure TUR (Mettler Toledo FSC402).

Total suspended solids (TSS) were measured using the filtration method, then drying the filtered sample at 105°C. Total nitrogen (TN as N) was measured using the Kje1dahl method. Ion chromatography (Dionex DX-120) was used to analyze chloride, nitrate, phosphate, and sulfate. Potassium and sodium were determined by a flame photometer (Jenway Clinical PFP7). Calcium was measured by ethylenediaminetetraacetic acid (EDTA) titrimetric method, and magnesium was measured by the difference between calcium and hardness (APHA, 2005).

HCO_3 was determined using the titration method. Equation (1) was used to calculate the sodium adsorption ratio (SAR), as described by Lesch and Suarez (2009); Inductively Coupled Plasma (ICP-OES - Perkin Elmer, Model 2000 DV) was used to measure heavy metals.

$$SAR = \frac{[Na^{+}]}{\sqrt{\frac{[Ca^{2+}]+[Mg^{2+}]}{2}}} \quad (1)$$

The BOD-5Day method was used to determine biological oxygen demand (BOD5), and the Closed Reflux, Titrimetric Method was used to measure chemical oxygen demand (COD) (APHA, 2005).

For microbiological analysis, water samples were collected in sterilized glass bottles with sodium thiosulphate. The samples were kept cold in an icebox below (10°C) during the transport period. The multiple tube fermentation method (MTF) was used to count *E. coli* and total *Coliforms* (APHA, 2005). The modified-Bailenger method was used to count *Helminth* eggs (Ayres et al., 1996). *Salmonellae* were measured as described by APHA (2005) and (Collee et al., 1989).

2.3. Soil analyses

Potatoes were harvested in April 2021, after 104 days of planting. Soil samples were collected from each plot, between the plants (emitters) for each depth of 0-20 cm, 21-40 cm, and 41-60 cm. Three soil samples were homogenized as composite samples from each plot for each depth. The air-dried soil samples were crushed and sieved using a 2-mm screen.

The soil texture was determined using the pipette method (Gee and Bauder, 1986); The oven-dry reference mass was used to calculate field capacity using the gravimetric method; the bulk density of the soil was determined using the core technique (Blake and Hartge, 1986). Equation(2) was used to calculate porosity. According to Richards (1954), the saturation paste extract and soil suspension was prepared.

$$Porosity = 1 - \left(\frac{BulkDensity}{ParticleDensity}\right) \quad (2)$$

ECe (dS m^{-1}) and pH were measured (1 soil: 1 water) as described by Richards (1954) and Jackson (1958), respectively. Sodium was measured directly by flame photometer according to Richard (1954). The sodium adsorption ratio (SAR) was calculated according to equation (1); Kjeldahl digestion was used to measure total nitrogen, then by distillation of steam (Jackson, 1958); the hot water technique was used to measure boron (B) (Gupta, 1993).

The Walkley-Black method was used to determine organic carbon (Walkley and Black, 1934). Organic matter (OM) % =Total Organic Carbon (TOC) % × 1.729. The DTPA method (Lindsey and Norvell, 1978) was used to determine micronutrients such as manganese (Mn), iron (Fe), copper (Cu), and zinc (Zn), using atomic absorption spectrophotometer (AAS) (Perkin Elmer, model AA 300). *E. coli* and total *Coliforms* were analyzed by the multiple tube method according to Turco (1994). The MPN table was used to calculate the most probable number (MPN) according to Cochran (1950). The results were expressed in MPN/g.

2.4. Yield and microbiological characteristics

Potatoes were harvested after four days of the last irrigation event, from the middle rows of each treatment. Potatoes were collected, weighed, and enumerated for every plant.

For microbial analysis, composite samples (six medium-sized fruits, around 500 g) were collected from the middle rows of each treatment. Gloves were changed to prevent contamination between plots. To detect *E.coli*/total *Coliform*, 500 ml sterile of 0.1% buffered peptone water (BPW) aliquots were added to a nylon bag containing the vegetable sample, as described by Seow et al. (2012). To suspend the microorganisms from the surface of the fruits, the sample was rubbed and shaken for 2 minutes in a nylon bag to catch the microorganisms present on the surface of the fruit. For each appropriate dilution (1: 10 of the rinse fluid was prepared using BPW), the sample (0.1 ml) was spread on chromogenic agar (Brilliance *E. coli*/*Coliform*; Oxoid), following a 24-hour incubation time at 37°C. Pink and violet colonies have been counted. Results were reported as colony-forming units per gram (cfu/g).

Salmonellae were measured by suspending the microorganisms from the surface of the fruits as described above (Seow et al., 2012). Then, filtering the sample through a 47 mm and 0.45 um (HA membrane filter, Millipore Corp), as described by APHA (2005). The filter membrane was then thoroughly blended with 100 ml of sterilized BPW (0.1 %), and then the sample was selectively enriched. The 0.1 ml of samples were streaked after enrichment and Biochemical and serological tests were used to confirm the isolates (colonies), according to Collee et al. (1989). The results were reported as colony-forming units per gram (cfu/g).

Helminth eggs were detected by preparing homogenate BPW as described above, then, the modified-Bailenger method was used to count *Helminth* eggs (Ayres et al., 1996).

2.5. Statistical analyses

Treatment effects were determined using analysis of variance (ANOVA). When the F ratio was significant, the Tukey-Kramer HSD was used to compare the mean values of all parameters at a 0.05 probability level. Statistical analyses were performed with the program JMP software (Version 12, 2015, SAS Institute Cary, NC, USA) (SAS, 2015). Deviation from the mean is presented in the tables of water quality as standard deviation. The relationship between potatoes number and their weight in each treatment was estimated using the Linear Regression JMP model (SAS, 2015)

3. Results and discussion

3.1. Irrigation water characteristics

Chemical and biological analyses were done to assess CIW and TWW (Table 1). Most of the average characteristics of TWW and CIW used for irrigation (in mg L^{-1}) were within the limits of FAO recommended concentrations (Pescod, 1992), and the technical regulation of reclaimed domestic wastewater use, number 893/2021 in Jordan (JSMO, 2021). The turbidity of TWW (25 NTU) was higher than the maximum limits for irrigation (10 NTU). High TUR and TSS in TWW could cause emitter clogging, particularly if micro-irrigation is used (Li et al., 2013; Pescod, 1992). The electrical conductivity (EC) of TWW (2.06 dSm^{-1}) was 3.27 times higher than CIW electrical conductivity (0.63 dSm^{-1}). In general, TWW salinity is 1.5 – 2 times higher than freshwater salinity, according to Chen et al. (2013).

Boron (B) content of CIW (2.08 mg L^{-1}) was 4.73 times higher than TWW (0.44 mg L^{-1}) and exceeded the maximum limits for irrigation in JS893/2021(JSMO, 2021) (1.0 mg L^{-1}) and FAO (Ayers and Westcot, 1985) (2 mg L^{-1}). Boron (B) is often found in high concentrations in association with saline soils and saline well water (Hilal et al., 2011). Higher B concentration can be interpreted as caused by distillation units of groundwater wells for CIW. The reverse osmosis (RO) membrane desalination process is an efficient and reliable technology for the production of drinking water from brackish water. However, RO membranes reject the B (Jung et al., 2020).

Potatoes are moderately sensitive to B (1.0 to 2.0 mg L^{-1}) (Pescod, 1992). The B is significant for improving potato tuber yield and quality by increasing the immunity of potato plants to early blight and thus reducing the usage of fungicides in crop production (Marschner, 2011).

Table (1) indicates that TWW contains higher amounts of total nitrogen (T-N), phosphorous ($P\text{-}PO_4$), and potassium (K), as compared with the CIW, which are necessary for plant growth and development. In Jordan, wastewater can supply about 75% of the fertilizer needs of typical farms (Carr et al., 2011). Heavy metals including micronutrients (Fe, Mn, Cu, and Zn), nickel (Ni), chromium (Cr), cadmium (Cd), and lead (Pb) were measured in irrigation water and were within acceptable values.

On the other hand, microbial pollution is one of the significant issues, which is directly related to the health risks of using TWW for agricultural irrigation. In terms of total *Coliforms*, *E. coli*, *Salmonella*, and *Helminth* eggs, the microbiology quality of water was assessed. No microorganisms were detected in the CIW, while the mean concentrations of *E. coli* in TWW (5.7×10^4 MPN 100 ml^{-1}) were greater than the limit (100 MPN 100 ml^{-1}) required for irrigating vegetables according to JS 893/2021. *Salmonella* and *Helminth* eggs were absent in TWW (Table 1). These findings agreed with several studies that found no *Salmonella* in municipal TWW (Lonigro et al., 2016; Cirelli et al., 2012). The presence and/or concentrations of the most important pathogens in water cannot be accurately predicted by only coliform indicators (Pachepsky et al., 2016).

The absence of *Salmonella* could indicate that the treated water eco-environment is harsher, more complex, and more dynamic. Numerous environmental conditions influence Salmonella's ability to survive and persist in water (Wanjugi and Harwood, 2013). In addition, the removal of suspended solids in WWTP aids in the control of pathogenic organisms and viruses and makes disinfection more effective. Because disinfectants such as chlorine and ozone react with organic compounds, thus pathogens become protected from disinfectants (Winward et al., 2008). These results reflect the treatment effectiveness of Kufranja WWTP or the low prevalence of Salmonella infections in the community. These results agreed with the findings reported by Karpiscak et al. (2001), which indicated that the much higher turbidity of wastewater the lesser effectiveness of treatment systems to remove some microbial indicators and pathogens.

Table 1. Characteristics of irrigation water in the study.

Parameters	Conventional irrigation water (CIW)		Treated wastewater (TWW)		Jordanian Standards (JS 893/2021/Class 1)
ECw (dSm^{-1})	0.63	±0.05	2.06	±0.53	2.3
pH	7.69	±0.12	7.60	±0.16	6-9
Cl^-($mg\ L^{-1}$)	97.82	±2.67	202.91	±88.76	400
SO_4^{-2} ($mg\ L^{-1}$)	58.62	±4.87	79.89	±16.88	500
HCO_3^-($mg\ L^{-1}$)	57.96	±4.76	351.89	±134.89	400
$P\text{-}PO_4^{-3}$ ($mg\ L^{-1}$)	0.00	±0.01	18.69	±11.40	30
$N\text{-}NO_3^-$($mg\ L^{-1}$)	3.52	±2.60	12.02	±3.18	30
K^+ ($mg\ L^{-1}$)	6.11	±1.26	47.44	±10.52	N/A
$B\text{-}H_3BO_3$($mg\ L^{-1}$)	2.08	±0.15	0.44	±0.07	1.0
Ca ($mg\ L^{-1}$)	20.57	±1.72	91.67	±19.52	230
Mg^{+2} ($mg\ L^{-1}$)	23.97	±3.49	37.57	±8.57	100
Na^+ ($mg\ L^{-1}$)	74.22	±10.87	200.56	±27.02	230
SAR	2.39	±0.78	4.46	±0.29	9.0
T-N ($mg\ L^{-1}$)	-	-	9.92	±1.60	N/A
TSS ($mg\ L^{-1}$)	-	-	41.70	±12.80	50
TUR (NTU)	-	-	25	±27.00	10
BOD5 ($mg\ L^{-1}$)	-	-	25.44	±4.44	30
COD ($mg\ L^{-1}$)	-	-	59.92	±9.01	100
DO ($mg\ L^{-1}$)	-	-	4.09	±3.58	>2
Cu^{+2} (ppm)	< 0.008	-	< 0.008		0.2
Fe^{+3} (ppm)	< 0.013	-	0.06	±0.00	5.0
Zn^{+2} (ppm)	< 0.017	-	< 0.017	-	5.0
Mn^{+2} (ppm)	< 0.017	-	< 0.017	-	0.2
Cd^{+2} (ppm)	< 0.009	-	< 0.009	-	0.01
Cr^{+2} (ppm)	< 0.005	-	< 0.005	-	0.1
Ni^{+2} (ppm)	< 0.01	-	< 0.01	-	0.2
Pb^{+2} (ppm)	< 0.008	-	< 0.008	-	0.2
TC ($MPN100mL^{-1}$)	< 1.1	-	>1600000	-	N/A
E. coli ($MPN\ 100mL^{-1}$)	< 1.1	-	57333	±38911	100
Salmonella ($MPN\ L^{-1}$)	ND	-	ND	-	N/A
Nematode Eggs	ND		ND	-	≤ 1

ECw: electrical conductivity water; SAR: sodium adsorption ratio; BOD5: biochemical oxygen demand at 5 days; COD: chemical oxygen demand; TSS: total solid suspended; TIN: total inorganic nitrogen; DO: dissolved oxygen; NTU: N nephelometric turbidity units; TUR: turbidity; MPN: most probable number; CFU: colony-forming unit; ND: not detected.

3.2. Soil characteristics

3.2.1. Soil physical characteristics

The soil texture is primarily clay loam, silty loam, and clay loam at 0-20, 20-40, and 40-60 cm depths, respectively (Table 2). The clay loam soil has high field capacity, medium till ability, fair internal drainage, and low wind erodibility (Finkel, 2019). Deep, well-drained sandy loam soils to loam soils have the best characteristics for high-quality potato farming (Martins et al., 2018; Lambeth, 1953).

The results of soil porosity and bulk density after harvesting the crop are presented in Table (3). As expected, for a short period of TWW application, both soil porosity and bulk density were much more resistant to soil alteration by treated wastewater irrigation.

3.2.2. Soil chemical characteristics.

The impact of TWW irrigation on the soil's chemical properties is mainly reflected by the electrical conductivity (ECe) (Table 4). Soil salinity is undoubtedly a fundamental factor for soil suitability for crop production. Soil contains both organic and inorganic chemicals that contribute to salinity stress, such as $CaSO_4$, $MgCl_2$, NaCl, Na_2SO_4, $MgSO_4$, Na_2CO_3, and KCl (Strawn et al., 2020; Munns and Tester, 2008).

Table 2. Soil texture characteristics.

Soil separates	Soil depth (cm)		
	0 -20	21-40	41-60
Sand (%)	22.0	22.9	33.9
Silt (%)	42.3	53.2	35.2
Clay (%)	35.7	24.0	30.9
Texture	Clay loam	Silty loam	Clay loam

Table 3. Soil's porosity and bulk density characteristics after harvesting. (*).

Treatments	Porosity (%)	Bulk density (mg m^{3-1})
TWW	52.6 a	1.23 a
BIW	51.7 a	1.28 a
CIW	51.7 a	1.30 a

(*) Means with the same letters in the same column are not significantly different at the 0.05 probability level according to the Tukey-Kramer HSD test.

The results showed that soil ECe was significantly higher for treatment irrigated with TWW compared to treatments irrigated with BIW and CIW by 27% and 69%, respectively (Table 4). In addition, ECe for treatment irrigated using BIW was significantly higher than that for treatments irrigated using CIW by 33%. The significant increase in soil ECe in the TWW-irrigated plots (Table 4) resulted from the high concentration of salts in the TWW (2.06 dS m^{-1}) compared with CIW (0.63 dS m^{-1}). These results agreed with the findings of Kaboosi (2017) and Qadir et al. (2000).

The pH of the soil samples ranged between 7.73 and 7.86 (Table 4). The results showed no significant differences between treatments. Urbano et al. (2015) reported the same finding for five cycles of lettuce fields irrigated using TWW. These findings could indicate that the soil has a buffering effect, thus, the pH value is steady, particularly in clay or organic-rich soil (Masto et al., 2009).

Organic matter (OM) constitutes a significant part of the soil, and its content is routinely used to assess soil fertility (Mugo et al., 2020; Giusquiani et al., 1995). The SOM improves soil fertility, increasing water-holding capacity, and improving soil structure, plant productivity, and microbial activity (Masmoudi et al., 2020; Marinari et al., 2000). Soil OM plays a key role in global warming. As a result, sewage irrigation became one source of soil organic carbon in cropland, contributing to global carbon circulation (Rattan et al., 2005).

The results showed that TOC was significantly higher for TWW-irrigated plots compared with treatments irrigated with CIW by 38% (Table 4). Organic matter of TWW resulted in a significant increase in TOC in the TWW-irrigated plots. These results agreed with the previous findings reported by Bedbabis et al. (2014) and Rattan et al. (2005). Trost et al. (2013) reported a rise of 11% to 35% in soil organic carbon in semiarid regions, regardless of irrigation water type. The results showed that soil TN (Table 4), at the top layer (0-20 cm) was significantly higher for treatment irrigated with TWW compared with treatments irrigated with BIW and CIW by 15.7% and 29.6%, respectively. Whereas no significant difference in TN for the deeper depths (20-60 cm) between treatments. These results agreed with the findings reported by Guo et al. (2017) and Becerra-Castro et al. (2015).

The SAR was significantly higher with TWW-irrigated plots compared with treatments irrigated with CIW by 61% (Table 4). However, the results were below the level for soil to be classified as sodic. These results agreed with the findings reported by Bedbabis et al. (2014), Hentati et al. (2014), Sou et al. (2013), and Al-Hamaiedeh and Bino (2010). Petousi et al. (2019) studied the impacts of secondary using TWW irrigation, and their results revealed no significant differences in soil properties compared with control, except for SAR and the EC, which were slightly higher in TWW soil samples. Most of these studies attributed the high SAR to the salinity of TWW, limited rainfall, high evaporation rates, and lack of drainage infrastructure all contributing factors.

Table 4. Some soil chemical characteristics (*).

Treatments	Soil depth(cm)	ECe (dSm^{-1})	pH	TOC (%)	T-N (%)	SAR
TWW	0-20	1.15 a	7.82 a	1.34 a	0.140 a	4.7 a
BIW		0.71 b	7.77 a	1.16 ab	0.121 b	4.5 a
CIW		0.51 c	7.86 a	0.98 b	0.108 c	3.2 b
TWW	20-40	1.37 a	7.82 a	1.86 a	0.33 a	9.49 a
BIW		1.11 b	7.81a	1.47 ab	0.12 a	7.15 b
CIW		0.82 c	7.80 a	1.34 b	0.075 a	6.38 b
TWW	40-60	1.85 a	7.74 a	1.24 a	0.18 a	15.45 a
BIW		1.61 b	7.73 a	0.80 a	0.101 a	10.25 b
CIW		1.25 c	7.79 a	0.80 a	0.06 a	8.78 b

(*) Means with the same letters in the same column (are not significantly different at the 0.05 probability level, according to the Tukey-Kramer HSD test.

Soil micronutrients are important to plant growth, plants need small amounts of them (Marschner, 2011). They are generally higher in the topsoil and decrease with soil depth. The most important soil micronutrients include boron (B), iron (Fe), copper (Cu), zinc (Zn), and manganese (Mn). Micronutrients can be a problem in case of their high concentration in the soil over time and phytotoxic bioaccumulation (Atafar et al., 2010).

Soil B was significantly higher for treatment irrigated with CIW compared with treatments irrigated with TWW and BIW by 54% and 35% (Table 5), respectively. The higher concentration of B in the CIW (2.08 mg L^{-1}) causes a significant increase in soil B compared to TWW (0.44 mg L^{-1}). Irrigation water is a key cause of excessive levels of soil B (Zaman et al., 2018), but some of the excess B could be leached by rainfall and/or irrigation. However, because B is adsorbed on soil particles, leaching could be problematic, and much higher water is needed. Soils in arid regions may have naturally toxic levels of B (García-Sánchez et al., 2020), making TWW use more difficult

The Fe concentrations were significantly higher with TWW-irrigated plots in the soil surface layer compared with treatments irrigated with CIW by 36.2% (Table 5). The higher concentration of Fe in the TWW (0.06 mg L^{-1}) caused a significant increase in Fe in the TWW-irrigated plots compared to CIW (<0.013 mg L^{-1}). The Mn concentrations were significantly higher with TWW-irrigated plots in the soil's deeper layer (40-6 cm) compared with treatments irrigated with CIW by 75%. (Table 5), while no significant differences were observed in the upper layers (Table 5). These results could be influenced by fertilizer applications made before the experiment. According to Wuana and Okieimen (2011), heavy metal traces could be found in N, P, and K fertilizer compounds.

The TWW irrigated soil (Zn and Cu) content was almost the same within all irrigation treatments. These results differ from other studies that showed soil heavy metal concentrations increased with TWW irrigation. Khaskhoussy et al. (2015) reported that TWW irrigation increased copper concentration in the soil. In another study, Fe and Zn increased a two-to eight-fold accumulation in the soil surface after two years of irrigation using TWW (Salgado-Méndez et al., 2019). On the other hand, several studies showed significantly increased heavy metals concentration in clayey soil irrigated with TWW compared with sandy soils (Alnaimy et al., 2021; Kinuthia et al., 2020; Hidri et al., 2014; Klay et al., 2010). Diverging findings can be interpreted, that heavy metal levels in domestic wastewater in Jordan that are used for irrigation of crops were within the recommended levels by the world standards as reported by Othman et al., (2021), as well as Jordan is not an industrial country, and industrial wastewater is treated separately. Furthermore, the low loading rate during irrigation with such water contributes to the slow accumulation of heavy metals (Abdelrahman et al., 2011; Mohammad and Mazahreh, 2003). Nevertheless, the long-term use of this reused water could increase the risk of contaminating soil and crops with several toxic heavy metals.

3.2.3. Soil microbial characteristics.

Pathogens are the main health concern and the most common threat to wastewater reuse in agriculture, both for workers and consumers. However, other sources of contamination, such as stray animals and birds, have the potential to contaminate the irrigated soil with *E. coli*, as reported by Fonseca et al., (2020) and Venglovsky et al. (2006). The most microbial reliable indicator for water reuse in irrigation, is the *E. coli* count, and the most important indicator that shows the potential presence of harmful bacteria causing diseases (Price and Wildeboer, 2017; Pescod, 1992).

The *E. coli* was not detected, while the total *Coliform* was 7 MPN g^{-1} in the soil before the beginning of the experiment. García-Orenes et al. (2007) reported that the decrease in soil water content under semiarid conditions could be the main factor in the decrease of *Coliform*. After harvesting, total *Coliform* and *E. coli* were significantly higher in TWW irrigated plots compared to CIW plots, where total *Coliform* and *E. coli* increased from 16 to 803 and from 1.8 to 120 (MPN g^{-1}) (Table 6), respectively. These results agreed with the findings reported by Petousi et al. (2019), Farhadkhani et al. (2018), Al-Rashidi et al. (2013), and Gerba and Smith (2005).

3.2.4. Yield and microbiological characteristics.

(1) Yield: Several crops have been successfully irrigated with TWW (Maaß and Grundmann, 2018; Hanjra et al., 2012), with crop yields increasing from 10% to 30% (Lazarova and Bahri, 2004). The marketable yield of tomatoes in the TWW application was 1.21 times greater than the value of the FW application, according to Demir and Sahin (2017).

Table 5. Soil chemical characteristics after harvesting (*).

Treatments	Soil depth (cm)	B (ppm)	Cu (ppm)	Fe (ppm)	Zn (ppm)	Mn (ppm)	Ni (ppm)
TWW	0-20	2.02 b	3.96 a	7.57 a	6.58 a	5.43 a	2.14 ab
BIW		2.2 b	3.39 a	6.61 a b	5.38 a	5.51 a	2.70 a
CIW		2.85 a	2.64 a	5.56 b	4.49 a	5.02 a	1.91b
TWW	20-40	1.51 b	1.75 a	5.85 a	4.0 a	4.33 a	3.17 a
BIW		1.80 b	1.39 a	6.40 a	2.9 a	4.40 a	2.91 a
CIW		2.4 a	1.54 a	5.80 a	3.2 a	2.80 a	2.25 a
TWW	40-60	0.62 b	1.22 a	5.75 a	4.13 a	4.65 a	2.70 a
BIW		0.75 b	1.10 a	6.65 a	2.70 a	5.00 a	1.65 a
CIW		1.14 a	0.67 a	6.25 a	3.65 a	2.65 b	2.13 a

(*)Means with the same letters in the same column (depth) are not significantly different at the 0.05 probability level, according to the Tukey-Kramer HSD test.

Table 6. Soil microbial characteristics after harvesting (*).

Treatments	Total coliform (MPNg^{-1})	E. coli (MPNg^{-1})
TWW	803 a	120 a
BIW	190 b	26 b
CIW	16 c	1.8 c

(*) Means with the same letters in the same column are not significantly different at the 0.05 probability level according to the Tukey-Kramer HSD test.

In this study, potato harvesting started after four days of stopping irrigation. The results showed that potato yield increased significantly for treatment irrigated with TWW (30.7 tons ha^{-1}) compared with treatments irrigated with CIW (25.7 tons ha^{-1}) by 19.5% (Table 7). Potatoes yield in TWW irrigated plots was higher than that in BIW irrigated plots by 6%, even though it was not significantly different. In addition, potatoes yield in BIW-irrigated plots was higher than that in CIW - irrigated plots by 12.7%. These results agreed with the findings of potatoes experiments reported by Abdul Mojid and Wyseure (2014), Marofi et al. (2013), and Zavadil (2009)

In this study, the results showed that the number of potatoes increased significantly for treatment irrigated with BIW compared with treatments irrigated with TWW and CIW by 6% and 21.9%, respectively (Table 7). The relationship between the increase in the number of potatoes for each treatment, and the increase in their total weight, was estimated using the Linear Regression JMP model. The results showed that the increase in total weight (kg) was significantly higher for treatment irrigated with TWW compared with treatments irrigated with BIW and CIW (Figure 1).

Table 7. Impact of irrigation water quality on potatoes yield (*)

Treatments	Yield (ton ha^{-1})	Yield (fruits ha^{-1})
TWW	30.70 a	117,600 ab
BIW	28.96 ab	124,800 a
CIW	25.70 b	102,400 b

(*) Means with the same letters in the same column are not significantly different at the 0.05 probability level, according to the Tukey-Kramer HSD test.

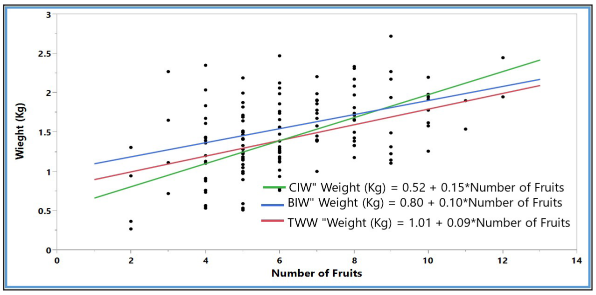

Figure 1. The relationship between the increase in the number of potatoes and the increase in their total weight for each treatment. Significant at a level of P<0.05.

On the other hand, it is natural that the lower the number of fruits per plant, the greater the weight per fruit. Figure 2 shows the relationship between the decrease in the number of potatoes per plant and the increase in weight per fruit for each treatment. The results showed the average increase in weight per fruit was significantly higher for treatment irrigated with TWW compared with treatments irrigated with BIW and CIW.

These results agreed with the findings reported by several authors (Pedrero et al., 2018; Nicolás et al., 2016; Pedrero et al., 2012). Pedrero et al. (2018) found that the size of nectarine fruit increased as the number of fruits per plant decreased, using TWW. In addition, the irrigation with TWW significantly reduced the number of nectarine fruits per tree compared to freshwater. However, the increase in the size of the fruits compensated for the reduction in the number of fruits.

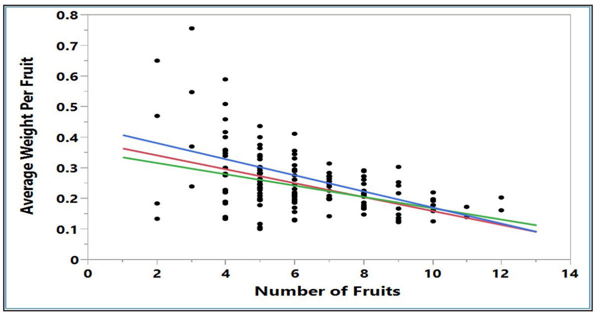

Figure 2. The relationship between the decrease in the number of potatoes per plant and the increase in weight per fruit for each treatment. Significant at a level of P< 0.05.

(2) Microbiological quality

Crops irrigated with TWW may carry pathogens such as parasitic, viral, and bacterial diseases to consumers (Domenech et al., 2018; Spanakos et al., 2015; WHO, 2006). Farhadkhani et al. (2018) reported that the irrigation method and plant type could be the most significant factors in crop microbial contamination.

Microbiologically, the harvested potatoes were analyzed. Total *Coliforms* were significantly higher on the surface of potato tubers in the TWW irrigated plots compared with BIW and CIW, (560, 70, and 30 cfu g^{-1}, respectively) (Table 8). No significant increase in *E. coli* was observed between treatments. These results agreed with those reported by Battilani et al. (2014); they observed no significant increase in *E. coli* for potatoes irrigated with TWW.

Pathogen indicators, such as *Salmonella* and *Helminth* eggs, were not found (Table 8). Lonigro et al. (2016) reported the same results. Chen et al. (2013) reported that there is limited evidence of the spread of the disease using TWW for agricultural irrigation. In contrast, *E. coli* recorded a significant increase for radishes under the drip and furrow system, but *Salmonella* was absent (Bastos and Mara, 1995). The presence of *E. coli* in the CIW irrigated plots could be attributed to different sources of contamination, such as roaming animals, and birds (Venglovsky et al., 2006). The contrast in the different studies' results could be because of the difference in the TWW quality, environment, or management method.

The successful measures reduce the potential microbial contaminations by reduction of the exposure of workers to wastewater. Some of the measures, such as drip irrigation and stopping irrigation before harvest, could play a significant role in the successful use of TWW for irrigation. A period without irrigation before harvest (1-2 weeks) can allow the die-off of bacteria and viruses to improve the quality of irrigated crops to levels seen in crops irrigated with fresh water, as reported by Vaz da Costa Vargas et al. (1996). However, this option is workable for some crops such as potatoes, and unworkable for vegetables that need harvesting daily, because farmers will probably not wait or stop irrigation of leafy salad crops five days or more before harvest (Lamm, 2002; Aiello et al. 2007).

Table 8. Potatoes' microbiological characteristics.

Treatment	Total *coliform* (cfu g^{-1})	*E.coli* (cfu g^{-1})	*Salmonella* (cfu g^{-1})	*H.*eggs (egg g^{-1})
TWW	560 a	40 a	ND	ND
BIW	70 b	20 a	ND	ND
CIW	30 b	10 a	ND	ND

(*) Means with the same letters in the same column are not significantly different at the 0.05 probability level, according to the Tukey-Kramer HSD test

4. Conclusions

Water reuse is essential in Jordan. The TWW contributes significantly to the country's limited irrigation water supply, allowing agriculture to continue in some areas. The study showed the effects of irrigation with TWW on some physical and chemical properties of the soil. Physically, in the short term, no significant differences were observed. Chemically, in particular, TWW increased the organic matter and TN, specifically in the surface soil. Regarding the SAR and EC, TWW induced a significant accumulation in the soil layers. The higher B concentration in soil for CIW irrigated plots is due to the higher concentration of B in irrigation water. Microbially, total *Coliform* and *E. coli* were significantly higher in TWW irrigated plots compared with CIW plots. On the other hand, potatoes showed significant contamination by total *Coliform*. Therefore, although non-significant observed of *E.coli* and the absence of pathogens such as *Salmonella* and *Helminth* eggs, root and tuber crops may be exposed to microbial hazards. Thus, a period of stopping irrigation should be scheduled before harvest.

Finally, the results of this study indicated that the use of TWW with drip irrigation for potato production could be feasible with TWW. Another key fact is that TWW and BIW treatments tended more considerable fruit weight and size. However, more research is needed to show the positive impacts of TWW on potatoes' fruit quality measures.

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Disclosure statement

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