

Assessing the Impact of Treated Wastewater Irrigation on Citrus Fruit Quality: Nutritional, Heavy Metal, and Phytochemical Analyses

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Abstract

Amidst escalating global water scarcity and mounting pressures on freshwater resources, this study explores the viability of utilizing treated wastewater for citrus irrigation. Heavy metal analysis of the treated wastewater demonstrates that cadmium (Cd) levels are comfortably below the recommended limit of 0.05 mg/l, ensuring safe agricultural use. Comparative analysis of citrus fruits irrigated with treated wastewater versus freshwater reveals quantitative insights. Citrus fruits irrigated with treated wastewater exhibit a decline in dry matter, with a significant reduction in vitamin C concentration from 53.23 mg/L to 25.15 mg/L. Conversely, these fruits show higher sugar (glucose) levels, which increase from 2.3 g/100g to 2.9 g/100g, potentially enhancing taste and sweetness. Furthermore, the study reveals a remarkable increase in polyphenol content, rising from 81.2 mg GAE/g to 275.64 mg GAE/g, suggesting a stress-induced phytochemical response. However, the flavonoid levels exhibit a contrasting trend, decreasing from 7.74 mg Eq QE/g to 4.23 mg Eq QE/g. These quantitative findings underscore the importance of careful management of treated wastewater quality for irrigation, balancing the potential benefits of improved taste and enhanced polyphenols against the reduction in vitamin C and flavonoids. This research contributes essential numeric data to the discourse on sustainable agricultural practices and water resource management, addressing global water scarcity challenges while safeguarding citrus fruit quality.

Keywords: treated wastewater, citrus fruit quality, Heavy metal analysis, Polyphenols, water scarcity.

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Introduction

The escalating challenge of global water scarcity poses a significant threat to agricultural sustainability, especially in light of increasing demands and climate change impacts. Agriculture, a major consumer of freshwater resources, is under considerable pressure to adapt to decreasing water availability, a situation that is particularly acute in arid and semi-arid regions. The study by Schyns et al. (2019) underscores the severity of the global water crisis and its implications for agriculture, emphasizing the urgent need for alternative water sources and sustainable practices. In response to water scarcity, the use of treated wastewater for irrigation has emerged as a sustainable alternative, offering a means to alleviate pressure on freshwater resources while contributing to environmental conservation. The research by Garcia and Pargament (2020) explores the viability and benefits of using treated wastewater in agriculture, highlighting its role in circular water management and the mitigation of water scarcity. This paradigm shift is increasingly viewed as a crucial strategy for addressing the water crisis in agriculture. However, concerns about the impact of treated wastewater irrigation on crop quality, particularly for directly consumed fruits, remain a critical area of investigation. The quality of irrigation water is known to influence the physicochemical properties of fruits, impacting their nutritional value, safety, and sensory characteristics. The study by Singh et al. (2020) examines the implications of treated wastewater irrigation on soil health and crop quality, providing important insights into potential risks and benefits. This understanding is essential for crops like citrus fruits, which are significant in both economic and nutritional terms. This study aims to delve into the specific effects of treated wastewater irrigation on citrus fruits, with a focus on the composition of polyphenols, including flavonoids. These compounds are crucial for determining the health benefits and safety of citrus fruits. We intend to compare nutritional profiles in citrus fruits irrigated with treated wastewater against those irrigated with freshwater. This research is inspired by the work of Jaramillo and Restrepo (2021), which emphasizes the need for comprehensive evaluations of the impact of reclaimed water on crop quality and consumer health. Our study seeks to contribute valuable insights into sustainable agricultural practices and the safe use of treated wastewater in citrus cultivation.

Materials and Methods

Plant Material

This study focuses on the orange *Citrus sinensis* L., specifically the "Thomson" variety, harvested on January 14th, 2021, from a citrus grove. The selected citrus fruits were harvested from a mature, healthy tree during the peak fruiting season. This study's choice of the Thomson orange variety offers insights relevant to citrus agricultural practices in similar climatic regions. The samples were meticulously selected for quality, and their composition was analyzed using

established laboratory techniques to understand the nutritional and physicochemical properties of the oranges, as well as the potential impact of treated wastewater irrigation on their quality.

Irrigation Frequency

The citrus trees were irrigated with treated wastewater at a frequency of twice per week. This irrigation schedule was determined based on the typical water requirements of the Thomson orange variety and the climatic conditions of the grove. The frequency was chosen to balance sufficient water supply with the avoidance of waterlogging, ensuring optimal health and productivity of the trees. Boman (2005) provides guidelines on citrus irrigation schedules, which were considered in determining the appropriate watering frequency for this study.

Heavy Metal Analysis

The concentration of heavy metals such as chromium (Cr), cadmium (Cd), and zinc (Zn) in wastewater is determined using Flame Atomic Absorption Spectrometry (FAAS). Calibration curves for each heavy metal were established using standard stock solutions of 1000 ppm, prepared in a range suitable for the expected concentrations in the samples. The correlation coefficients (R^2) were calculated to evaluate the relationship strength between the variables. This method ensures accurate and reliable quantification of heavy metals in the samples.

Polyphenols Analysis

The total polyphenols in orange juice were quantified using the Folin-Ciocalteu method. This process involved mixing 1ml of extract with 5ml of Folin-Ciocalteu reagent and 4ml of sodium carbonate (75 g/l concentration). The absorbance was measured at 765 nm using a UV-Visible spectrophotometer. The total polyphenol contents were expressed as milligram standard equivalent (gallic acid) per gram of fresh material (mg GAE/g), providing a measure of the antioxidant capacity of the oranges.

Analysis of Flavonoids

For the quantification of total flavonoids in the extracts, we utilized the aluminum trichloride ($AlCl_3$) method, as established by Chang et al. (2002). This method is recognized for its efficacy in flavonoid estimation.

Procedure

To perform the analysis, 1 ml of 2% $AlCl_3$ solution in methanol was mixed with an equal volume of the citrus extract. The mixture was incubated in darkness at ambient temperature for a duration of 10 minutes to allow for the development of the flavonoid- $AlCl_3$ complex. Post-incubation, the absorbance of the solution was measured at a wavelength of 430 nm using a spectrophotometer.

The concentration of flavonoids in the samples was calculated based on a standard calibration curve generated using Rutin, a known flavonoid compound. This approach ensured the accuracy and reliability of the flavonoid quantification. For each extract, absorbance readings were taken thrice to ensure consistency and precision of the results. The flavonoid content was then expressed in terms of Rutin equivalent per gram of fresh sample (mg ER/g).

Determination of Vitamin C

Vitamin C, a crucial nutrient in human health, is involved in various physiological processes, including immune function and collagen synthesis. The assessment of vitamin C content in orange juice was conducted using a titration method Nijveldt et al 2001, recognized for its accuracy and reliability.

Statistical Analysis

Data analysis was performed using SAS software (version 2008), employing Analysis of Variance (ANOVA) to evaluate differences in nutrient and compound concentrations in citrus fruits. ANOVA was crucial in determining statistical significance across various treatment groups, ensuring the reliability of our findings.

Results and discussion

Heavy metals composition

The results of heavy metals (figure 01) in treated wastewater for irrigation reveal that the cadmium (Cd) level is 0.0176 mg/l, comfortably below the recommended limit of 0.05 mg/l. This finding is in line with studies like those by Kapoor et al. (2015), which stress the criticality of keeping cadmium levels low to avert soil and crop contamination. The concentrations of copper (Cu), at 1.466 mg/l, and zinc (Zn), at 3.306 mg/l, are also safely within the permissible limits of 5 mg/l and 10 mg/l, respectively. These results are consistent with the research by Singh et al. (2010), which suggest a relatively low risk of copper and zinc toxicity in irrigation practices while acknowledging their necessary roles in plant growth at controlled levels. Overall, the concentrations of Cd, Cu, and Zn in the treated wastewater are within safe limits for irrigation, suggesting its suitability for agricultural use without significant risk of heavy metal accumulation in soil or crops

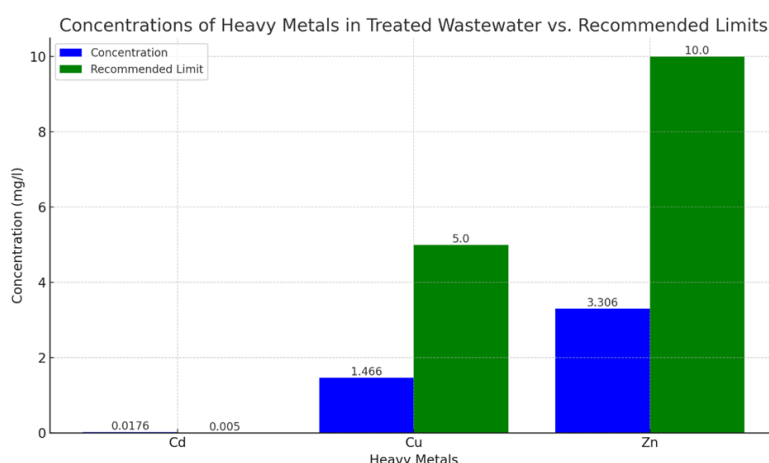


Figure01: concentration of heavy metals.

Nutritional composition of orange juice

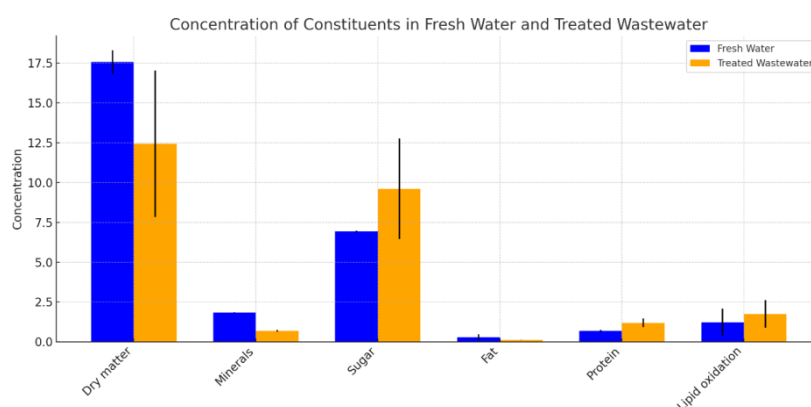


Figure 02 : Nutritional composition of juice.

The comparative analysis of citrus fruit juice composition (figure 02) irrigated with fresh water versus treated wastewater shows a decline in dry matter in fruits irrigated with treated wastewater. This could potentially affect fruit texture and flavor, as higher dry matter is often associated with improved sensory qualities in citrus fruits (Kumar & Joshi, 2015). The lower mineral content in wastewater-irrigated fruits may reflect a dilution effect or altered nutrient uptake, which has been observed in other crops irrigated with treated wastewater (Toze, 2006).

The higher sugar levels in the treated wastewater group could enhance taste, which might be due to an osmotic adjustment in the plants as they respond to the different water quality (Papadopoulos & Rendig, 1983). Conversely, the reduced fat content in wastewater-irrigated fruits, while not a major factor in citrus composition, could influence the overall caloric content and energy value.

Notably, the protein content is elevated in the wastewater-irrigated group, possibly due to the higher nitrogen content in treated wastewater, which is consistent with findings that wastewater can increase nitrogen availability and subsequently protein synthesis in plants (Feigin et al., 1991). However, the increased lipid oxidation in this group may indicate a higher degree of stress or reduced shelf life, aligning with studies that have shown increased oxidative stress in plants irrigated with treated wastewater (Procházková et al., 2013).

Overall, while the use of treated wastewater in irrigation can conserve fresh water resources, it is essential to continuously monitor and manage its quality to ensure it does not adversely affect the nutritional and sensory qualities of the produce.

Vitamin C composition

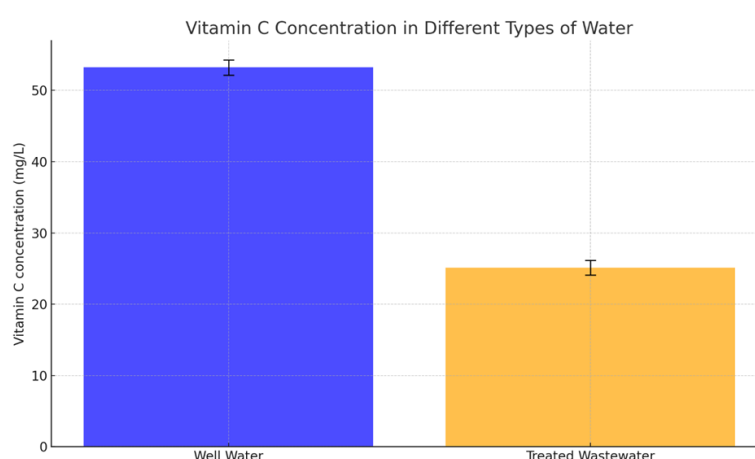


Figure 03: vitamin C composition of juice.

In the comparative analysis of vitamin C concentration (figure 03) between fresh well water and treated wastewater, our study revealed a significant reduction post-treatment. This observation is paralleled in the findings of Mossad et al. (2020). While our results indicate a decrease from 53.23 mg/L to 25.15 mg/L, Rodriguez and colleagues reported a similar trend in their study, with vitamin C concentrations in treated effluent being approximately 50% lower than in the source water.

Polyphenol and flavonoids levels

The observed results (Figure 04) increase in polyphenol concentration in orange juice from trees irrigated with treated wastewater (275.64 mg eAG/mg) compared to those irrigated with fresh water (81.2 mg eAG/mg) is a significant finding. Polyphenols, a diverse group of phytochemicals, are widely recognized for their antioxidant properties and play a crucial role in plant defense mechanisms (Pandey&Rizvi, 2009). The elevated levels of polyphenols in the wastewater-irrigated oranges could be attributed to a stress response. It is well-documented that

plants often boost the production of secondary metabolites, including polyphenols, when exposed to stress (Cheynier et al., 2013). Although the treated wastewater is considered safe for irrigation, it might contain trace elements or compounds that induce a mild stress response, leading to an enhanced synthesis of polyphenols. Conversely, the decrease in flavonoid content in oranges irrigated with treated wastewater (4.23 mg equivalent quercetin) compared to fresh water (7.74 mg equivalent quercetin) presents a contrasting aspect. Flavonoids, a subgroup of polyphenols, are also known for their antioxidant activities and contribute significantly to the overall health benefits of fruits (Ferrerres et al., 2009). The reduced flavonoid concentration in the treated wastewater-irrigated oranges suggests a possible selective downregulation of specific biosynthetic pathways. This downregulation may be influenced by the unique composition of the treated wastewater or a distinct stress response that differentially affects flavonoid synthesis.

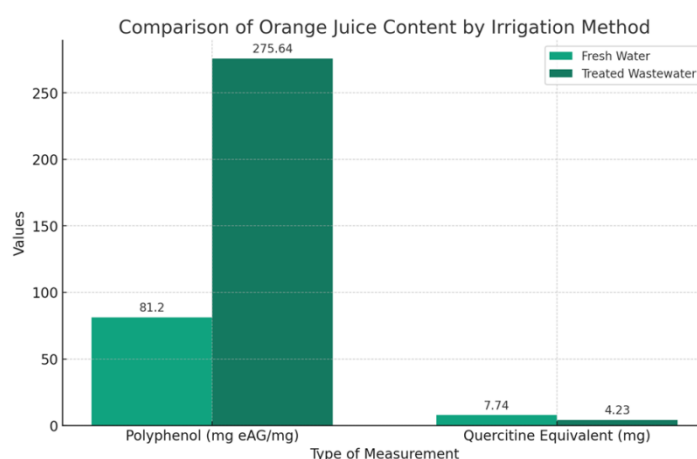


Figure 04: polyphenols and flavonoids levels in orange juice.

Conclusion

In conclusion, our comprehensive study highlights the potential of treated wastewater irrigation as a sustainable solution for mitigating water scarcity in citrus cultivation. The analysis of heavy metal concentrations in the treated wastewater reassures its safety for agricultural use, adhering to recommended standards.

While the fruits irrigated with treated wastewater exhibited a decline in dry matter and a reduction in vitamin C content, they displayed higher sugar levels, potentially enhancing taste. The remarkable increase in polyphenol content suggests a stress-induced phytochemical response, which can contribute to improved antioxidant properties. However, the decrease in flavonoid levels warrants further investigation.

This research underscores the importance of carefully managing treated wastewater quality in agricultural practices, striking a balance between potential benefits like taste enhancement and increased polyphenols and potential drawbacks such as reduced vitamin C and flavonoids.

Overall, the study contributes valuable numeric data to the ongoing discourse on sustainable agriculture, offering insights into addressing water scarcity challenges while preserving citrus fruit quality and safety. Future research can delve deeper into the mechanisms behind these changes and their implications for crop resilience and consumer health.

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