Enhancing the Valorization of Secondary Treated Wastewater Irrigation for Seed Germination and Early Seedling Growth of Tomato (Solanum lycopersicum L.) Using Moringa oleifera Leaf Extract under Greenhouse Conditions

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Abstract
This study investigated the potential of Moringa oleifera leaf extract (MLE) to enhance the agronomic performance of secondary treated wastewater (TWW) used for seed germination and early seedling growth of tomato (Solanum lycopersicum L. cv. Marmande) cultivation. A Petri-dish germination assay and a pot experiment conducted under greenhouse conditions were carried out. Tomato seeds were irrigated with TWW alone or with TWW supplemented with MLE at 2, 4, and 6 mL L^{-1}. Germination behaviour, early seedling growth, morphological traits, and physiological parameters, including chlorophyll content and relative water content, were evaluated. MLE application significantly improved germination rate (GR) and uniformity, with the most pronounced effects observed at moderate concentrations (2–4 mL L^{-1}). These improvements were associated with enhanced seedling vigour, increased chlorophyll accumulation, and better plant water status. In contrast, the highest concentration (6 mL L^{-1}) slightly reduced these benefits, indicating a dose-dependent biostimulant response. Overall, combining MLE with treated wastewater represents an eco-efficient strategy to improve crop establishment while mitigating the constraints associated with reclaimed water use in arid and semi-arid agriculture.

Keywords: treated wastewater; Moringa oleifera; tomato; plant biostimulant; seed germination; sustainable agriculture.

Introduction

Water is a fundamental natural resource that underpins socio-economic development and sustains agricultural and industrial activities worldwide. Its availability strongly influences settlement patterns, agricultural productivity, and long-term regional development strategies (Bkhaita & Fraj, 2025). However, increasing population growth, climate variability, and competing sectoral demands have intensified pressure on conventional freshwater resources, making sustainable water management a critical global challenge (Carrière, 2024). Water scarcity currently affects more than 50 countries, particularly those located in arid and semi-arid regions of the Mediterranean Basin (Ruslana et al., 2025).

Algeria is among the countries most severely exposed to chronic water scarcity. Renewable freshwater resources are limited to only a few billion cubic meters per year, with nearly two-thirds concentrated in the northern regions (Guereueb & Ferhat, 2013). Agriculture accounts for approximately 70% of total water withdrawals (Khemmouli, 2025), accelerating the depletion of resources that are already among the lowest per capita worldwide (Boukra, 2024). Annual rainfall ranges between 200 and 600 mm and exhibits strong spatial and temporal variability (Hallouz et al., 2013). These constraints, combined with low soil fertility and challenging topography, limit agricultural intensification, despite the strategic importance of agriculture for national food security in an economy still largely dependent on hydrocarbon revenues (Bakhtach et al., 2023). In 2020, per capita water availability in Algeria was estimated at only 411 m^3 per year, well below the absolute scarcity threshold of 500 m^3 per year (Kherbache, 2020).

Under these conditions, the development and valorisation of non-conventional water resources, particularly treated wastewater (TWW), have become essential components of sustainable water management strategies. The reuse of TWW offers multiple advantages, including reduced abstraction of freshwater resources, nutrient recycling, and the maintenance of crop production under limited water availability (López et al., 2019). Several studies conducted in Algeria have reported that effluents from wastewater treatment plants, such as the Aïn Sefra facility, comply with FAO irrigation standards, exhibiting stable physicochemical characteristics and effective nitrate removal (Moussaoui et al., 2023). Nevertheless, despite the existence of a supportive legal framework (Kouider et al., 2025), less than 0.2% of treated wastewater is currently reused for agricultural irrigation in Algeria (Bendida, 2024). This low adoption rate highlights persistent technical, agronomic, and environmental constraints related to water quality, salinity, nutrient imbalance, and potential microbial risks.

In recent years, biostimulant-based approaches have emerged as promising tools for improving plant performance under abiotic stress while supporting sustainable agricultural practices (Christou et al., 2024). Among natural biostimulants, Moringa oleifera has attracted considerable attention due to its multifunctional properties. Extracts derived from Moringa leaves or seeds are rich in phytohormones, vitamins, antioxidants, and phenolic compounds that enhance plant growth, physiological performance, and stress tolerance. For instance, foliar application of Moringa seed extract improved water productivity, chlorophyll content, and antioxidant activity in pepper (Capsicum annuum) under deficit irrigation (Buthelezi et al., 2022), while leaf extracts enhanced growth, photosynthetic pigments, and seed quality in soybean (Glycine max) (Irshad et al., 2025).

Beyond its agronomic effects, Moringa oleifera has demonstrated high efficiency as a natural coagulant in water treatment, achieving substantial reductions in turbidity and chemical oxygen demand (Mahaveerchand et al., 2024). Compared with conventional chemical coagulants, Moringa-based agents are biodegradable, non-toxic, and cost-effective, making them particularly suitable for water-scarce and developing regions. Recent studies further suggest that Moringa-derived coagulants can contribute to producing irrigation-grade water while limiting pollutant accumulation and the dissemination of antibiotic resistance genes (Sané et al., 2024; Gutiérrez-Herrera et al., 2024).

However, despite these advances, little information is available on the combined use of treated wastewater and Moringa oleifera leaf extract during the critical early stages of crop establishment, particularly seed germination and early seedling growth. These stages are highly sensitive to osmotic, ionic, and microbial stresses associated with reclaimed water irrigation, which can compromise plant establishment and subsequent productivity.

Therefore, the objective of this study was to evaluate whether the incorporation of Moringa oleifera leaf extract into treated wastewater irrigation could improve germination, early seedling growth, and physiological performance of tomato (Solanum lycopersicum L.). By exploring the synergistic potential of natural biostimulants and treated wastewater, this work contributes to the sustainable valorisation of non-conventional water resources and the development of resilient agricultural systems in arid and semi-arid environments.

Materials and Methods

Water collection

Treated wastewater (TWW) was collected from the Béni Merrad wastewater treatment plant (36°31′45″ N, 2°52′13″ E; **Fig.1**) located in Blida Province, Algeria. The facility spans approximately 6 ha and has a treatment capacity of 383,000 population equivalents (p.e.). It includes both wastewater and sludge treatment lines designed to produce purified effluent suitable for agricultural reuse. The plant operates an activated-sludge process providing secondary-level treatment; the effluent used in this study therefore corresponds to secondary treated wastewater. The plant receives a mixture of domestic and industrial wastewater from the surrounding catchment area. After treatment, the effluent that is not used for irrigation purposes is discharged into a natural watercourse.

Figure 1. Aerial view of the Beni Merrad wastewater treatment plant (Blida, Algeria), showing the wastewater and sludge treatment units (Original, 2025).

Preparation of Moringa oleifera leaf extract (MLE)

Leaves of 12-year-old Drumstick tree were collected in 2023 from Oued El Alleug (Blida Province, Algeria). The leaves were washed with distilled water, air dried in the dark for 10 days, and ground into a fine powder using an electric grinder. The powdered material was macerated in 70% ethanol (1:40, w/v) for 72 h at room temperature (25 ± 2 °C) with occasional stirring **(Vongsak et al., 2013). The extract was filtered through Whatman No. 1 paper, and the filtrate was diluted with distilled water in order to obtain final concentrations of** 2, 4, and 6 mL L^{-1} for experimental application.

2.3 Plant material and experimental design

Tomato (Solanum lycopersicum Linnaeus, 1753), cultivar 'Marmande', an indeterminate-growth cultivar suitable for both fresh-market consumption and processing, a widely cultivated Algerian variety of high agronomic value, was used as the test species. Seeds were obtained from the Technical Institute for Horticultural and Industrial Crops (ITCMI, Algiers). Prior to sowing, seeds were surface sterilized in 2% sodium hypochlorite for 10 min and rinsed thoroughly with sterile distilled water. Two separate assays were conducted as follows:

Petri dish assay: Seeds were placed on moistened filter paper in sterile Petri dishes and incubated at 25 °C in an incubator, with ten seeds per dish and four replicates per treatment (40 seeds per treatment). Treatments consisted of TWW alone or TWW supplemented with MLE at the designated concentrations. Germination percentage, kinetics, and early seedling traits (hypocotyl and radicle length) were recorded.

Pot assay: The pot experiment was conducted under greenhouse conditions. Seeds were sown in plug trays filled with sterilized peat substrate and kept in darkness until cotyledon emergence. Uniform seedlings were transplanted into 3 L plastic pots containing sterilized expanded clay (5–8 mm diameter).

During the first two weeks, seedlings received a standard nutrient solution with the following ionic composition (meq L^{-1}): NO_3^- 10.20, SO_4^{2-} 1.50, PO_4^{3-} 3.30, Cl^- 0.60, H^+ 3.30, Na^+ 1.30, K^+ 4.25, Ca^{2+} 5.10, Mg^{2+} 1.80, NH_4^+ 1.80. The pH was adjusted to 5.66, and electrical conductivity (EC) maintained at 2.11 mS cm^{-1}. A volume of 100 mL of this nutrient solution was applied per pot at each application.

After acclimatization, irrigation treatments were applied three times per week for one month in a completely randomized design (CRD) with four replicates per treatment, using a fixed volume of 100 mL per pot at each irrigation event:

Table 1. Description of irrigation treatments combining treated wastewater (TWW) and Moringa oleifera leaf extract (MLE).

Code	Treatment description
T0	TWW without MLE (control)
T1	TWW + 2 mL L^{-1} MLE
T2	TWW + 4 mL L^{-1} MLE
T3	TWW + 6 mL L^{-1} MLE

Water quality analysis

Before application, TWW samples were analyzed for physical, chemical, and microbiological characteristics following the Standard Methods for the Examination of Water and Wastewater (APHA, AWWA & WEF, 2012).

- Physical parameters: pH, temperature, electrical conductivity (EC), turbidity, and residual chlorine.
- Chemical parameters: NO_3^-, NO_2^-, NH_4^+, PO_4^{3-}, SO_4^{2-}, Cl^-, Ca^{2+}, Mg^{2+}, K^+, Na^+, Fe, and Mn.
- Microbiological parameters: total coliforms, Escherichia coli, enterococci, and sulphite-reducing bacteria.

Germination and early growth measurements

Germination rate (GR): The percentage of germinated seeds was calculated according to Côme (1970) as the ratio of germinated seeds to the total number sown.

Germination kinetics: The number of germinated seeds was recorded at regular intervals, and cumulative germination (%) over time was plotted to evaluate both rate and uniformity.

Hypocotyl and radicle length: Measured at the end of the germination period using a digital caliper (precision ± 0.01 mm) to assess early seedling vigor (Gairola et al., 2011).

Morphological traits

Root and Shoot Length:

Root length was measured using a graduated ruler, whereas shoot height was determined using a digital caliper (precision ±0.01 mm).

Leaf Number and Leaf Area

The number of fully expanded leaves per plant was recorded and leaf area (LA) was estimated using the empirical formula **LA=Length× width ×0.75,** originally proposed by Montgomery (1911). A correction factor of 0.75 is widely accepted for elliptical leaves, has been validated in several crops, including maize and tomatoes (Dwyer & Stewart, 1986; Keating & Wafula, 1992). Alternative coefficients (0.67–0.85) have been reported depending on leaf shape and species (Pandey & Singh, 2011; Zhen et al., 2018)."

Physiological parameters:

Leaf Water Content (WC, %)
Leaf water content was determined using the gravimetric method. Fresh leaf samples were immediately weighed to obtain the fresh weight (FW). The samples were then oven-dried at 70°C for 24 h until constant weight was reached and the dry weight (DW) was recorded. The water content (WC) was calculated as follows (Barrs & Weatherley, 1962):

$$\mathrm{WC}(\%) = \frac{\mathrm{FW} - \mathrm{DW} \times 100}{\mathrm{FW}}$$

Chlorophyll Content (SPAD Index)
Chlorophyll content in the outermost leaves was estimated using a portable SPAD meter (SPAD-502, Konica Minolta, Japan). Five readings were taken per plant at the mid-section of the leaf blade, and the mean value was calculated for each plant.
Statistical analysis
Data were subjected to one-way analysis of variance (ANOVA) with treatment as the main factor. Prior to ANOVA, normality of the residuals and homogeneity of variances were verified using the Shapiro–Wilk and Levene tests, respectively, to ensure that the underlying assumptions of parametric analysis were met. Mean comparisons were performed using Tukey's honestly significant difference (HSD) test at the 5% probability level. Statistical significance was considered at $p \leq 0.05$, with highly significant differences at $p < 0.01$. Analyses were conducted using SPSS software (version 22; IBM Corp., Armonk, NY, USA).

Results

Physicochemical and microbiological quality of treated wastewater.
The analysis of the treated wastewater (TWW) used for irrigation revealed that most physicochemical parameters were within the permissible limits defined by Algerian (N.A) standards for agricultural reuse. The measured values are presented in **Tab 2**.The water exhibited a slightly yellowish colour (9.3 mg L^{-1} Pt), acceptable odour and taste (dilution threshold = 4), neutral pH (7.2), and moderate turbidity (3 NTU). Electrical conductivity was 2001.5 µS cm^{-1}—well below the maximum permissible limit of 2800 µS cm^{-1}—indicating moderate salinity. Macronutrient concentrations such as potassium (10 mg L^{-1}) and calcium (150 mg L^{-1}) were within acceptable limits. However, ammonium (0.72 mg L^{-1}) and total phosphorus (6 mg L^{-1}) slightly exceeded their respective thresholds (0.5 mg L^{-1} and 5 mg L^{-1}). Similarly, oxidisability (6.5 mg L^{-1} O_2) was above the recommended limit of 5 mg L^{-1}. Nitrate (50 mg L^{-1}) reached the upper permissible boundary, while nitrite (0.15 mg L^{-1}) remained below its limit.
Heavy metals and potentially toxic elements, including iron (< 0.01 mg L^{-1}), manganese (0.04 mg L^{-1}), aluminium (0.1 mg L^{-1}), fluorides (1 mg L^{-1}), and cyanides (55 µg L^{-1}), were all within safe limits. From a microbiological perspective, TWW showed measurable bacterial contamination, with total coliforms (180 CFU 100 mL^{-1}), Escherichia coli (76 CFU 100 mL^{-1}), and enterococci (53 CFU 100 mL^{-1}) detected. These values exceed the Algerian regulatory threshold of 0 CFU 100 mL^{-1} for the irrigation of market garden (vegetable) crops, indicating that, from a strict regulatory standpoint, the TWW used in this study did not comply with national sanitary standards for this crop category. This non-compliance is likely attributable to residual disinfection limitations of the secondary (activated-sludge) treatment process, which is not specifically designed for complete pathogen removal, combined with the mixed domestic–industrial origin of the influent. Such residual microbial load represents a potential health risk for consumers, farm workers, and soil microbial communities if reclaimed water of this quality were applied to crops consumed raw. Complementary disinfection steps (e.g., UV treatment, chlorination, or membrane filtration) prior to agricultural use, together with restricting this effluent to crops not intended for raw consumption or to non-food applications, would be necessary to ensure regulatory compliance and safeguard public health.

Table 2: Physicochemical and microbiological characteristics of treated wastewater compared with national (N.A) and international (I.S) standards for agricultural reuse.

Parameter	Unit	Measured Value	Standard Limit (N.A / I.S)
Color	mg/L Pt-Co	9.3	15
Odor at 25 °C	Dilution threshold	4	4
Taste at 25 °C	Dilution threshold	4	4
Free residual chlorine	mg/L	0.06	>0.1
pH	pH units	7.2	6.5–9.0
Electrical conductivity at 25 °C	µS/cm	2001.5	2800
Temperature	°C	25	25
Turbidity	NTU	3	5
Ammonium (NH_4^+)	mg/L	0.72	0.5
Nitrites (NO_2^-)	mg/L	0.15	0.2
Phosphorus (P)	mg/L	6	5
Oxidizability	mg/L O_2	6.5	5
Calcium (Ca^{2+})	mg/L	150	200
Total hardness (as $CaCO_3$)	mg/L	350	500

Potassium (K^+)	mg/L	10	12
Chlorides (Cl^-)	mg/L	200	500
Sulfates (SO_4^{2-})	mg/L	130	400
Nitrates (NO_3^-)	mg/L	50	50
Iron (Fe)	mg/L	<0.01	0.3
Manganese (Mn)	mg/L	0.04	0.05
Aluminum (Al)	mg/L	0.1	0.2
Fluorides (F^-)	mg/L	1	1.5
Cyanides (CN^-)	µg/L	55	70
Total coliforms	CFU/100 mL	180	None
Escherichia coli	CFU/100 mL	76	0
Enterococci	CFU/100 mL	53	0
Sulfite-reducing bacteria	CFU/100 mL	Indeterminate	0

Seed germination

The influence of TWW and MLE supplementation on tomato seed germination is shown in **Fig 2**. Seeds irrigated with TWW alone achieved a final germination rate of 39.0 ± 7.9%, whereas the addition of MLE significantly enhanced this parameter, reaching 60.0 ± 0.0%, 76.7 ± 5.8%, and 75.7 ± 5.1% at 2, 4, and 6 mL L^{-1}, respectively. Germination kinetics demonstrated earlier and faster emergence under MLE treatments: by day 4, more than 70% of seeds had germinated under 4 and 6 mL L^{-1}, compared with less than 50% under TWW alone.

Seedling growth parameters followed a similar trend. Hypocotyl length increased from 74.7 ± 8.3 mm (control) to 127.3 ± 2.5 mm under TWW + 2 mL L^{-1} MLE, while radicle elongation improved from 32.3 ± 3.1 mm to 55.7 ± 3.2 mm under 4 mL L^{-1}, before declining slightly at 6 mL L^{-1} (43.3 ± 3.4 mm). Overall, MLE not only enhanced the final germination rate but also accelerated germination speed and improved seedling vigour. The 4 mL L^{-1} dose was optimal for germination percentage and radicle elongation, while 2 mL L^{-1} most strongly promoted hypocotyl growth, revealing a dose-dependent effect.

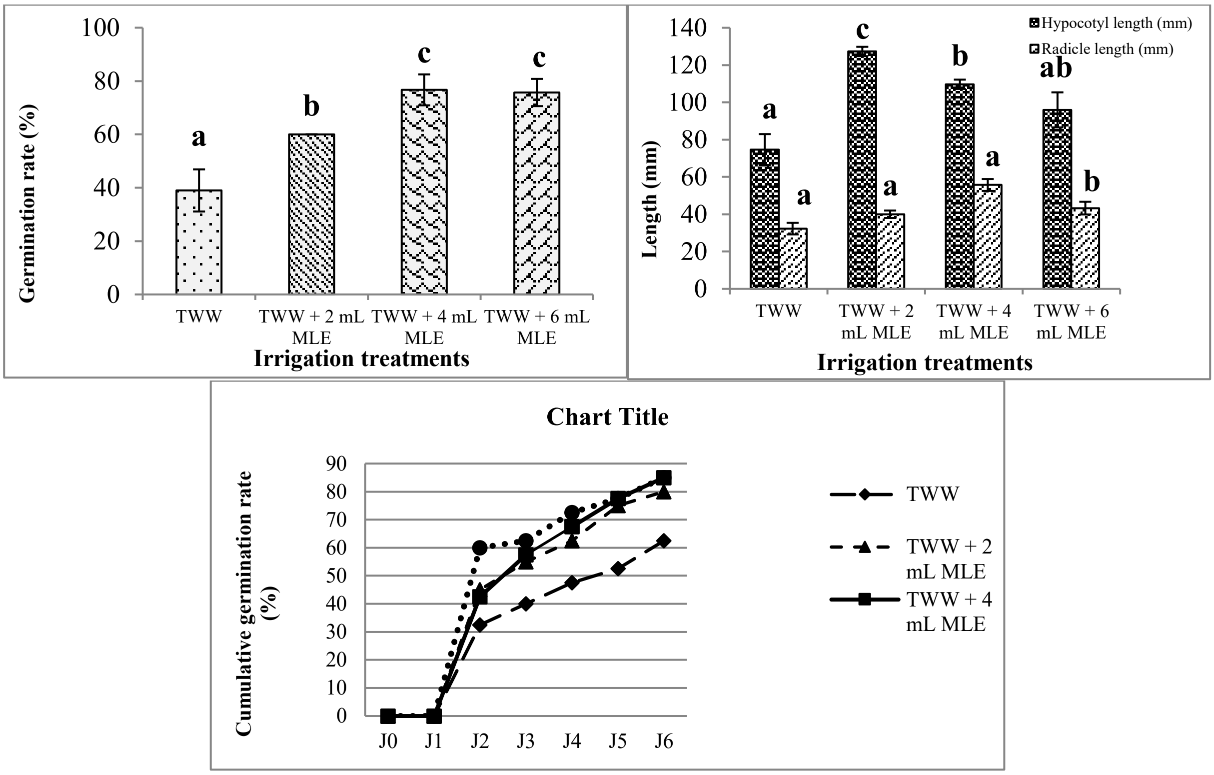

Figure 2. Effect of irrigation with treated wastewater (TWW) alone or combined with 2, 4, and 6 mL L^{-1} of Moringa oleifera leaf extract (MLE) on tomato seed germination rate, hypocotyl and radicle length, and germination kinetics.

(a) Germination rate (GR, %); (b) hypocotyl length (mm); (c) radicle length (mm); (d) cumulative germination kinetics (%) over time. Different letters indicate homogeneous groups according to ANOVA followed by Tukey's test ($p < 0.05$). Error bars represent standard deviation.

Morphological traits

As shown in **Fig3**, irrigation with TWW + MLE improved the morphological development of tomato seedlings compared with TWW alone. Stem elongation varied only slightly, from 18.2 ± 1.0 cm under 2 mL L^{-1} MLE to 21.0 ± 1.8 cm under 6 mL L^{-1}, compared with 19.8 ± 1.2 cm in the control. Root growth was more responsive, increasing from 12.5 ± 0.5 cm (control) to 15.5 ± 1.3 cm under 4 mL L^{-1} MLE.

Leaf development also improved markedly: the number of leaves rose from 14.7 ± 0.5 (control) to 16.3 ± 4.0 under 6 mL L^{-1}, and leaf area nearly doubled—from 8.8 ± 0.7 cm^2 (control) to 16.3 ± 1.8 cm^2 under 2 mL L^{-1} MLE. Overall, MLE supplementation enhanced seedling growth in a dose-dependent manner, with 4 mL L^{-1} yielding the greatest root elongation and 2 mL L^{-1} inducing the largest leaf expansion, whereas higher doses mainly favoured leaf number and stem elongation.

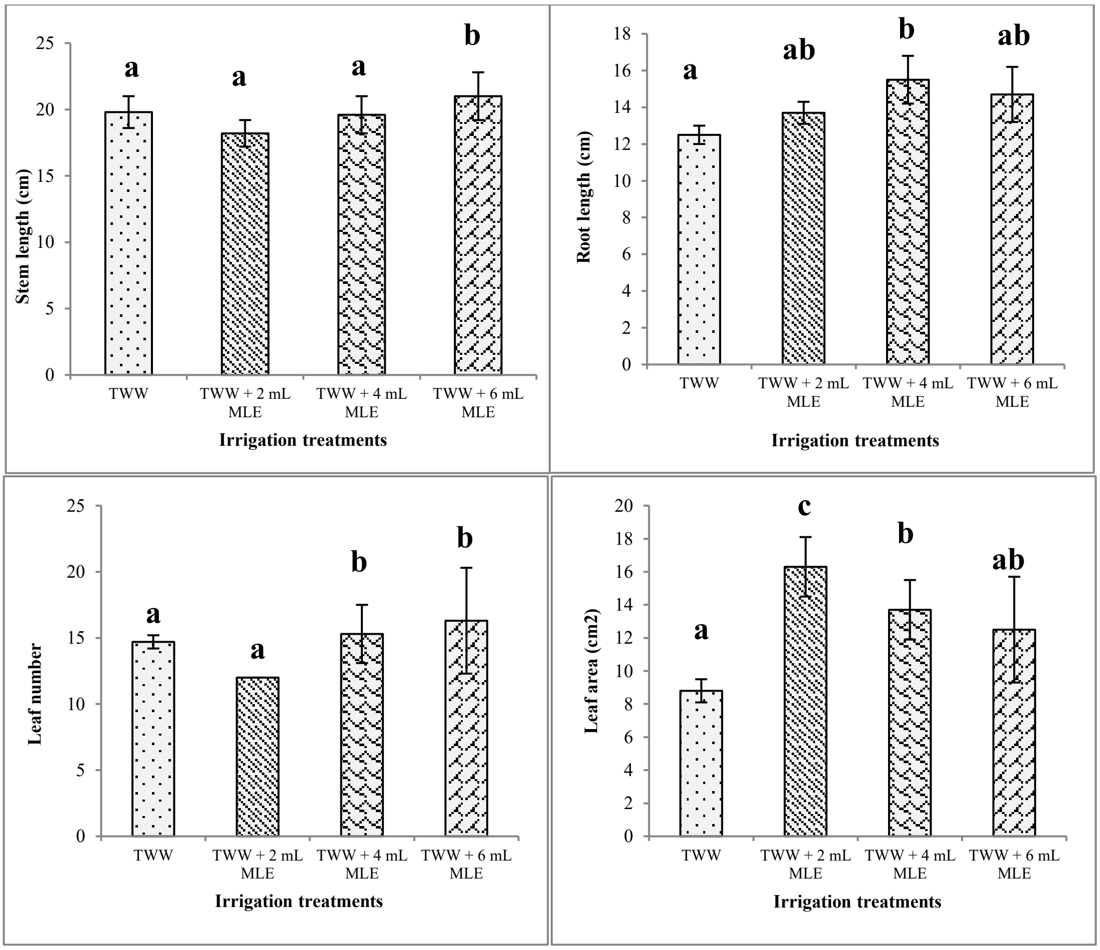

Figure 3. Effect of irrigation with treated wastewater (TWW) alone or combined with 2, 4, and 6 mL L^{-1} of Moringa oleifera leaf extract (MLE) on tomato seedling stem length, root length, leaf number, and leaf area.

(a) Stem length (mm); (b) root length (mm); (c) leaf number; (d) leaf area (cm^2). Different letters indicate homogeneous groups according to ANOVA followed by Tukey's test ($p < 0.05$). Error bars represent standard deviation

Physiological responses

The physiological performance of tomato seedlings under different treatments is presented in **Fig 4**. Chlorophyll content (SPAD index) increased from 25.9 ± 2.9 in the control to 37.0 ± 4.7 and 35.4 ± 1.2 under 2 and 4 mL L^{-1} MLE, respectively, but declined to 21.9 ± 3.5 under 6 mL L^{-1}. Similarly, relative water content (WC) improved from 63.2 ± 2.6% (control) to 85.5 ± 2.2% and 85.3 ± 1.9% under 2 and 4 mL L^{-1} MLE, before decreasing to 74.0 ± 2.8% at the highest dose.

These findings indicate that moderate concentrations of MLE (2–4 mL L^{-1}) enhanced photosynthetic pigment accumulation and water status in tomato seedlings, while excessive doses appeared inhibitory.

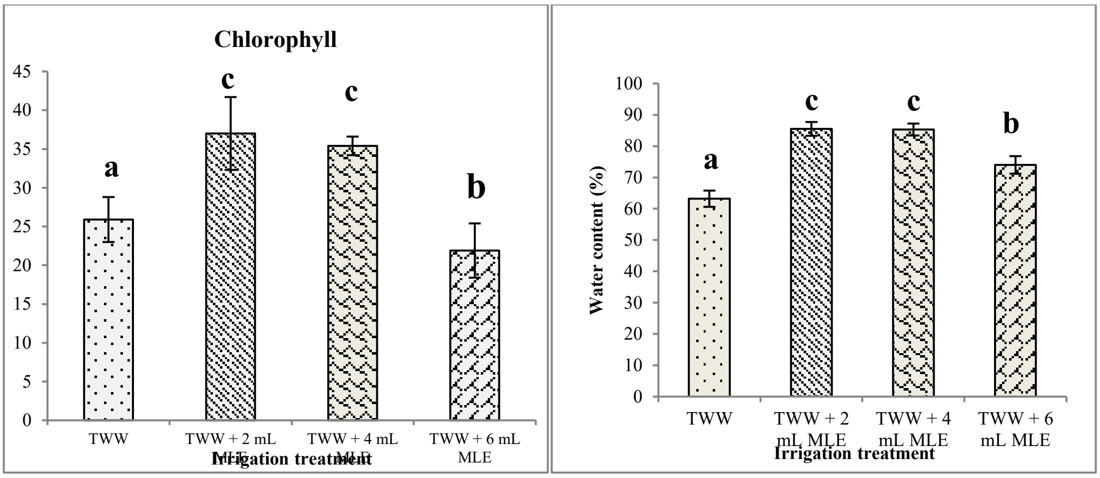

Figure 4. Effect of irrigation with treated wastewater (TWW) alone or combined with 2, 4, and 6 mL L⁻¹ of Moringa oleifera leaf extract (MLE) on leaf chlorophyll content (SPAD index) and leaf water content (%).

(a) Chlorophyll content (SPAD index); (b) leaf relative water content (%). Different letters indicate homogeneous groups according to ANOVA followed by Tukey's test ($p < 0.05$). Error bars represent standard deviation.

Discussion

Irrigation with treated wastewater (TWW) exposes seeds and seedlings to a combination of abiotic and biotic stressors, including osmotic imbalance, ionic toxicity, and microbial contamination. These constraints are known to promote excessive generation of reactive oxygen species (ROS), destabilise cellular membranes, and disrupt metabolic pathways essential for germination and early growth (Christou et al., 2024; Zhang et al., 2018). In the present study, irrigation with TWW alone markedly restricted tomato seed germination and seedling development. This inhibitory effect is likely associated with elevated ammonium and phosphorus concentrations and moderate microbial loads, despite the neutral pH and acceptable salinity of the effluent (Moussaoui et al., 2023).

The incorporation of Moringa oleifera leaf extract (MLE) into TWW substantially improved germination dynamics, seedling vigour, and overall growth performance. Moderate MLE concentrations (2–4 mL L^{-1}) enhanced germination speed and uniformity, hypocotyl and radicle elongation, root development, leaf number, and leaf area. In contrast, the highest dose (6 mL L^{-1}) resulted in reduced benefits, suggesting a hormetic dose–response pattern commonly reported for plant-based biostimulants (Biswas et al., 2016; Buthelezi et al., 2022; Irshad et al., 2025).

These improvements can be attributed to two complementary mechanisms. First, a hormonal mechanism, whereby phytohormones present in MLE—particularly cytokinins, auxins, and gibberellins—stimulate cell division, elongation, and chloroplast differentiation (Prasad, 2022). Second, an antioxidant and osmoprotective mechanism, as hydroalcoholic extracts of Moringa leaves contain phenolic and flavonoid compounds with strong free-radical scavenging capacity (Vongsak et al., 2013). Together, these processes promote chlorophyll accumulation, improve leaf water status, and enhance photosynthetic efficiency, thereby increasing tolerance to osmotic and oxidative stress induced by TWW irrigation. Enhanced root elongation and leaf expansion further indicate improved nutrient and water acquisition efficiency under these conditions.

The decline observed at the highest MLE concentration suggests that excessive application may induce hormonal imbalance or pro-oxidant effects linked to phenolic compound accumulation. Such bell-shaped responses are well documented in biostimulant studies and reflect the narrow optimal concentration range of bioactive compounds (Elzaawely et al., 2016; Prasad, 2022).

The present findings are consistent with recent studies reporting the stress-mitigating effects of Moringa-based biostimulants. For example, foliar application of MLE enhanced wheat growth and antioxidant enzyme activity under saline conditions (Rehman et al., 2024), supporting its role in alleviating oxidative stress. Similarly, improved maize growth and nutrient uptake under drought stress have been reported following Moringa extract application (Ali et al., 2025). Notably, unlike Ali et al. (2025), who observed sustained benefits at higher doses, our results indicate reduced efficiency at 6 mL L^{-1}, which may reflect differences in extraction protocols, application mode, or cultivar-specific sensitivity.

Under field conditions, combined use of TWW irrigation and biostimulants has been shown to enhance tomato yield and soil microbial activity (García-Martínez et al., 2024), suggesting that the positive responses observed here at early growth stages could translate into agronomic benefits at larger scales. However, environmental variability and long-term soil effects require careful assessment.

The interaction between TWW and MLE highlights a synergistic mechanism in which reclaimed water supplies essential moisture and nutrients but imposes physiological stress, while MLE mitigates these adverse effects by reinforcing antioxidant defence and physiological resilience. This synergy aligns with the principles of sustainable water reuse and circular agriculture (López et al., 2019; Kouider et al., 2025).

Nevertheless, the relatively short duration of this study and the limited replication may have constrained the detection of slower-developing traits, such as stem elongation and reproductive performance. Future research should therefore include field-scale trials assessing fruit yield, quality, and safety—particularly with respect to microbial contamination and heavy-metal accumulation—as well as soil salinity dynamics and long-term system sustainability. In addition, standardisation of extraction protocols and biochemical profiling of MLE (e.g. phenolics and phytohormones) are essential to improve reproducibility, given the influence of leaf maturity, provenance, and seasonal variation on extract composition (Vongsak et al., 2013).

Overall, the integration of Moringa oleifera leaf extract into treated wastewater irrigation emerges as a viable and eco-efficient strategy to enhance plant establishment and physiological performance in water-scarce environments, contributing to the sustainable valorisation of non-conventional water resources in Mediterranean and semi-arid regions.

Conclusions

Supplementation of treated wastewater (TWW) with Moringa oleifera leaf extract (MLE) significantly improved tomato germination and early seedling growth. Moderate MLE doses (2–4 mL L^{-1}) enhanced germination performance, seedling development, chlorophyll content, and plant water status. The combined use of TWW and MLE generated synergistic effects, mitigating stress associated with reclaimed water while promoting physiological and morphological growth. These results highlight the potential of Moringa oleifera as a low-cost biostimulant for the sustainable reuse of treated wastewater in semi-arid agriculture, although further field-scale studies are required to confirm its agronomic applicability.

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