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Effect of Nanopriming on emergence and seedling characteristics in parsley seeds

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Abstract: This study aims to prevent problems encountered by farmers during parsley cultivation and to determine the effectiveness of nanotechnology-based treatments, which are considered a new approach to sustainable agricultural practices. A total of 16 treatments were used in the study: a control and priming treatments (hydropriming, zinc priming, and zinc oxide nanopriming) with and without additives. After the treatments, the seeds were planted in the field. While the emergence rate of the control group was 18% at 45 days after planting, the highest emergence rate among treatments was 36% for the ferula-doped nanopriming treatments. Based on seedling measurements, it was observed that ferula- and magnesium-doped nanopriming treatments had a positive effect on seedling quality. When the yield calculation of the treatments under field conditions was performed, it was determined that the zinc oxide-doped, Ferula-doped, and magnesium-doped nanopriming treatments had the highest harvest efficiency.

Keywords: Sustainable agriculture, nano zinc oxide, green synthesis, *Petroselinum crispum*

Maydanoz tohumlarında Nanopriming uygulamalarının çıkış ve fide özelliklerine etkisi

Öz: Bu çalışma ile çiftçilerin maydanoz yetiştiricilik esnasında karşılaşmış oldukları problemlerin önüne geçmek, sürdürülebilir tarım uygulamalarında yeni bir yaklaşım olarak değerlendirilen nanoteknoloji uygulamalarının etkinliğini belirlemek hedeflenmektedir. Çalışmada kontrol, hidropriming uygulaması, çinko priming, katkılı ve katkısız çinkooksit nanopriming uygulamaları olmak üzere toplamda 16 farklı uygulama kullanılmıştır. Uygulamalar sonrasında tohumlar araziye ekilmiştir. Ekimden 45 gün sonra kontrol grubu çıkış oranı %18 iken ferula katkılı nanopriming uygulamasında %36 olmuştur. Ferula ve magnezyum katkılı nanopriming uygulamalarının fide kalitesini arttırdığı görülmüştür. Çinko oksit, ferula ve magnezyum katkılı nanopriming uygulamalarının maydanoz verimini arttırdığı belirlenmiştir.

Anahtar Kelimeler: Sürdürülebilir tarım, nano çinko oksit, yeşil sentez, *Petroselinum crispum*

Introduction

Parsley (*Petroselinum crispum*) is a crop of high economic value in Türkiye, particularly in the Hatay region. Hatay Province accounts for 60% of Türkiye's parsley production (TUİK, 2024). However, the small seed size can cause sowing problems, and the hard seed coat can lead to irregular and delayed germination or emergence. Seed germination and emergence are considered among the most critical stages in early agricultural production. The physiological state of the seed is the most important factor affecting germination and emergence. Low emergence rates and non-uniform seedling development, especially in open-field cultivation, are among the most significant problems farmers face. Another important factor affecting land emergence is suboptimal environmental conditions (e.g., temperature and rainfall). For these reasons, pre-sowing seed treatments are important for increasing the emergence

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rate and improving seedling development in parsley cultivation (Hassell and Kretchman, 1997; Dursun and Ekinci, 2010; de Oliveira et al., 2013; da Silva et al., 2022).

In recent years, nanopriming treatments have become increasingly popular as pre-sowing seed treatments due to their numerous positive effects on plant physiology, including increased germination and emergence rates, enhanced stress tolerance, improved seedling quality, activation of defense mechanisms under stress conditions, and increased enzyme content. Various synthetic methods are employed to obtain the agents used in nanopriming treatments. Nanomaterials can be produced using physical (top-down) techniques, chemical (bottom-up) techniques, green synthesis techniques, thermal techniques, and plasma-based techniques. While the production of nanomaterials through green synthesis supports sustainable and organic agriculture, the use of incorrect dosages in the production of nanomaterials through chemical synthesis can pose a threat to agricultural production. Furthermore, because nanomaterials produced through green synthesis are derived from plant extracts and natural biomolecules, they offer environmentally friendly applications in agricultural production, are safe for plant health, and offer low food residue risks. However, when applied at incorrect dosages, nanomaterials produced through chemical synthesis can pose risks such as residue risk, phytotoxicity, disruption of soil microbiota, and food safety risks. Therefore, green synthesis applications can be considered more advantageous in terms of sustainability in agricultural production (Özmen and Mavi 2023). The use of nanostructured agents developed by any of these techniques in nanopriming treatments relies on treating seeds with nanoparticle solutions at appropriate concentrations. Nanoparticles can positively influence the mechanisms of seed germination and emergence due to their high surface area, potential to regulate reactive oxygen species (ROS), and ability to facilitate ion exchange. Studies have shown that nanoparticles such as ZnO, Fe_3O_4, TiO_2, SiO_2, Ag, and CuO can increase germination rate, root and shoot development, enzyme activation, and stress tolerance when applied at appropriate doses in pepper, tomato, watermelon, bitter melon, and gourd species. Zinc is a vital micronutrient that plays roles in numerous physiological and metabolic processes and functions as a cofactor for many enzymes. Zinc oxide nanoparticles (ZnO-NPs) are becoming increasingly popular due to their superior efficiency, unique physicochemical, biological, and antimicrobial properties (Nile et al., 2022; Madusanka et al., 2024; Kalathingal and Palengara, 2025; Ochoa-Chaparro et al., 2025; Ranaware et al., 2025, Akkaya et al., 2024).

However, the mechanism of action may vary depending on the combination and dose of nanoparticles applied to each plant species (Dehkourdi and Mosavi, 2013; Nile et al., 2022). Therefore, determining the effectiveness of nanopriming treatments in species such as parsley, which experience germination and emergence problems, is important for agricultural production. This study aims to determine the physiological effects of nanopriming treatments on seedling development in parsley species and to contribute to developing sustainable and innovative approaches to pre-sowing seed treatments in seed technology. Therefore, the aim of this study is to determine the effects of different nanopriming treatments on developmental parameters such as emergence rate, seedling and root lengths, and seedling and root fresh and dry weights of parsley seeds under field conditions.

Materials and Methods

In this study, a standard variety of parsley (cv. Bezirci) was selected as the plant material, and seeds were supplied by the Balıkesir Seed Company. The seeds were stored at 4 °C until use in the study. A solution-based production technique was used to obtain the nanopriming agents for nanopriming treatments, thereby enabling the growth of nanostructured ZnO samples. Zinc acetate dihydrate ($Zn(CH_3COO)_2{\cdot}2H_2O$) was used to prepare solutions for the growth of ZnO samples. Ammonium hydroxide (NH_4OH) and distilled water were used as solvents to prepare the sample solution. The pH of the solution was then adjusted to approximately 10.0 using NH_4OH. After the powder fraction of the resulting solution had precipitated, the powder was filtered and annealed in an atmospheric oven at 250°C for 90 minutes, thereby yielding nanostructured ZnO powder. Nanostructured metal oxides are doped to enhance and tune their main physical properties for specific applications (Şahin et al., 2015, Şahin et al., 2019). Five different doping agents were used, and plant- and metal-based doping was applied to the ZnO structures at 10% by volume (Tasdemir et al., 2022, Figure 1).

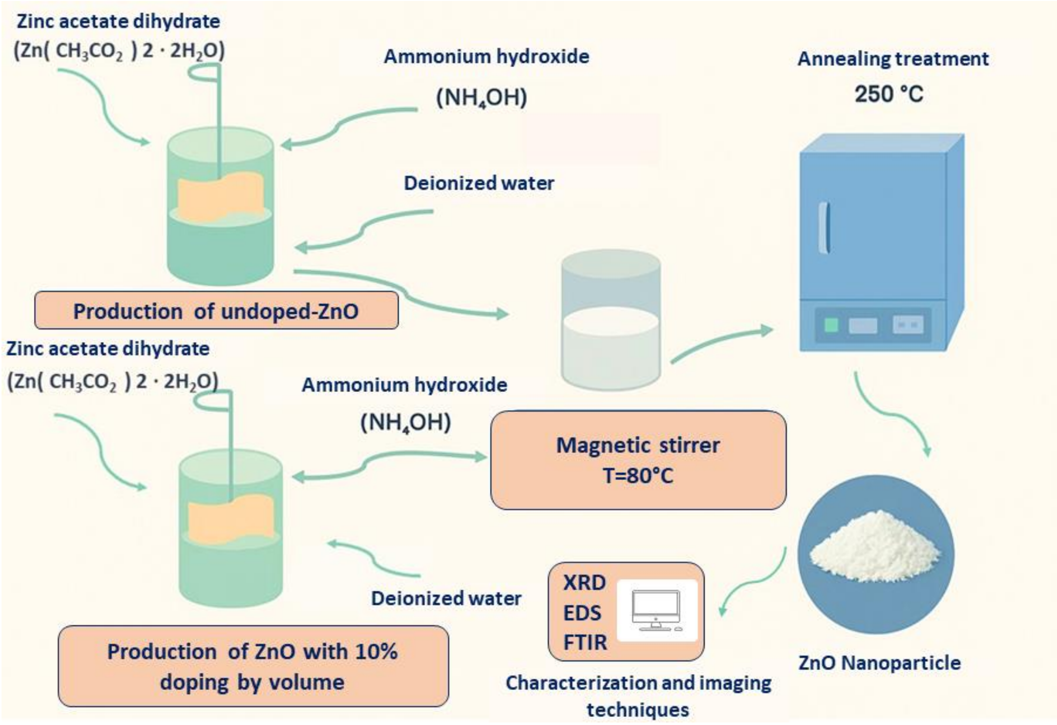

Figure 1. Production of selected additive and un-doped ZnO for use in nanopriming treatments

Whereas plant extracts obtained from Ferula and Tagetes were used as herbal additives, potassium, magnesium, and calcium salts were used as metal samples (Figure 2).

The herbal extracts were obtained by adding 0.05 g of Ferula gum to 250 mL of boiling water, the effective dose we determined in previous organic priming studies. For Ferula, we added 1 g of petals to 250 mL of boiling water. For *Tagetes erecta*, we added 1 g of petals to 250 mL of boiling water (Mavi, 2016; Mavi and Uzunoğlu, 2020).

A field-emission scanning electron microscope (FESEM), an energy-dispersive spectrometer (EDS), and an X-ray diffractometer (XRD) were used to examine the physical properties of the undoped and doped ZnO nanomaterials (Figure 1). Before establishing seedling emergence trials, seed lots were grouped into 3 × 50 replicates for use in control, hydropriming, and nanopriming treatments. Control-group seeds were separated and soweddirectly in the field without treatment. Seeds for hydropriming were placed in Petri dishes filled with 10 mL of deionized water and kept at 25°C for 24 hours. They were then dried to their initial moisture content and used in the seedling emergence trial.

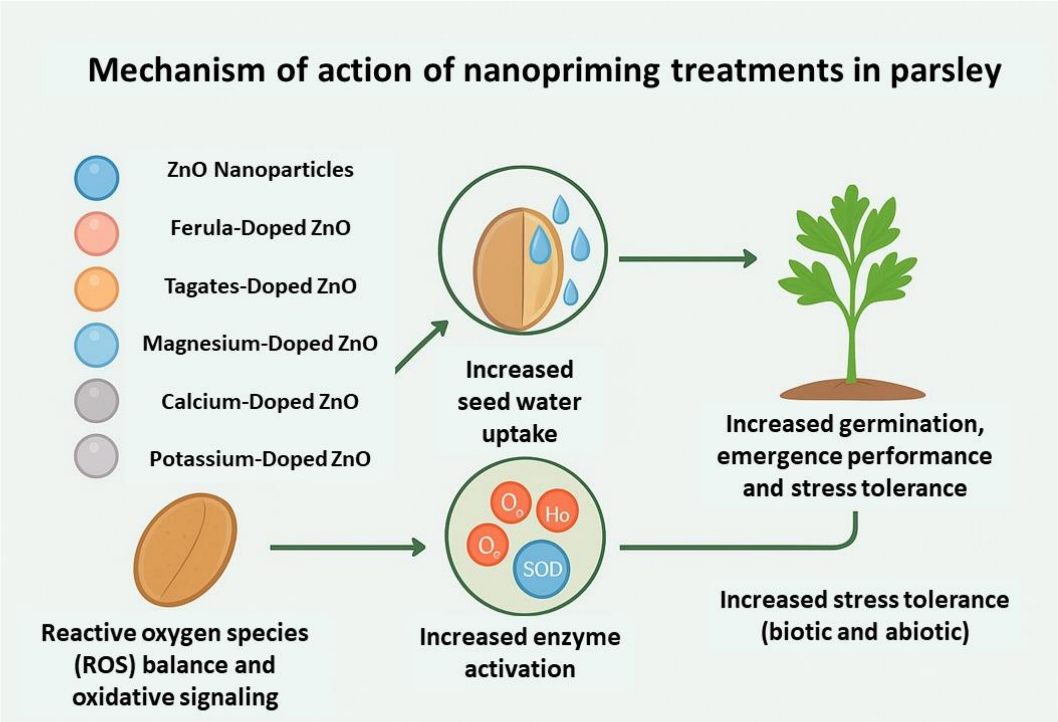

Figure 2. Nanopriming agents applied to parsley seeds and their mechanism of action

The nanopriming agents used in the study were nanostructured zinc oxide (ZnO), nanostructured tagetes-doped zinc oxide, nanostructured ferula-doped zinc oxide, nanostructured potassium-doped zinc oxide, nanostructured magnesium-doped zinc oxide, and nanostructured calcium-doped zinc oxide; these six agents were applied at two doses (50 and 100 ppm) (Özmen et al., 2022). A total of 16 treatments were studied (Table 1), including control, hydropriming, zinc, and nanopriming.

The nanopriming treatment was performed using 3 replicates of 50 seeds each placed between blotting papers in Petri dishes. To each dish, 10 mL of the treatment agent at the dose was added, and the dishes were kept at 25°C for 24 hours. At the end of this period, the seeds were dried to their initial moisture content and used in the seedling emergence trial. The seedling emergence trial was conducted at the Hatay Mustafa Kemal University field site, and seeds were sown in 3 × 50 replicates at a depth of 1 cm to ensure uniform planting depth. Distances of 10 cm and 20 cm were maintained between replicates and treatment groups, respectively.

Table 1. Treatments used in the study and their abbreviation codes

Treatments Name	Abbreviation code	Treatments Name	Abbreviation code
Control	M1	Ferula-doped ZnO NPs (50 ppm)	M9
Hydropriming	M2	Ferula-doped ZnO NPs (100 ppm)	M10
Zn (50 ppm)	M3	K-doped ZnO NPs (50 ppm)	M11
Zn (100 ppm)	M4	K-doped ZnO NPs (100 ppm)	M12
ZnO (50 ppm)	M5	Mg-doped ZnO NPs (50 ppm)	M13
ZnO (100 ppm)	M6	Mg-doped ZnO NPs (100 ppm)	M14
Tagetes-doped ZnO NPs (50 ppm)	M7	$Ca_3(PO_4)_2$-doped ZnO NPs (50 ppm)	M15
Tagetes-doped ZnO NPs(100 ppm)	M8	$Ca_3(PO_4)_2$-doped ZnO NPs (100 ppm)	M16

After emergence was completed, mean emergence rates were calculated as percentages. To determine changes in seedling quality 60 days after emergence was complete, seedling and root lengths (cm), stem and root diameters (mm), seedling (without roots) fresh and dry weights (g), root fresh and dry weights (g), leaf width (cm), and leaf length (cm) were measured on 3 replicates of 10 seedlings from each treatment group, and yield per decare (kg) was determined. The data obtained from the measurements were analyzed in SPSS software using Duncan's multiple-comparison test, and differences between treatments were determined at a significance level of $p<0.05$.

Findings and Discussion

According to the XRD results, the crystal structure of the synthesized nanomaterial was polycrystalline. When the peak heights and widths in Figure 3 are examined, the observed sharp, narrow peaks indicate that the obtained sample exhibits high crystallinity. This is also supported by the data from the JCPDS dataset [Reference code: 01-079-5249]. The predominance of the XRD peaks corresponding to the (100), (002), and (101) planes indicates that the produced sample is pure, single-phase ZnO (Figure 3).

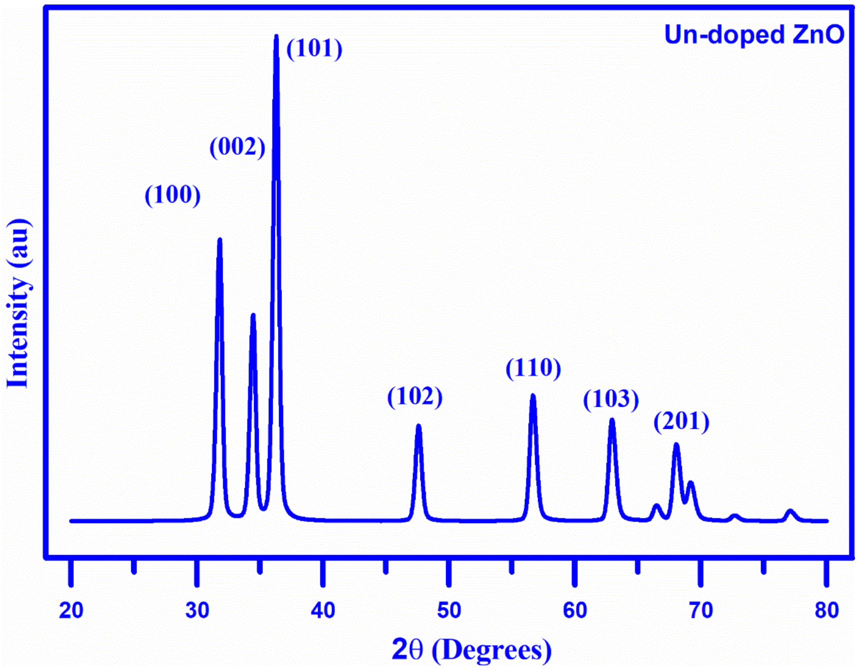

Figure 3. XRD patterns of un-doped ZnO nanostructures

The average crystal size was calculated using the Scherrer equation to be 17.50 nm for the undoped ZnO nanomaterial, 17.80 nm for the K-doped ZnO, 19.53 nm for the Mg-doped ZnO, 17.83 nm for the Ca-doped ZnO, 17.63 nm for the Ferula-doped ZnO nanomaterial, and approximately 16.84 nm for the Tagetes-doped ZnO nanomaterial (Table 2).

Table 2. Crystal sizes of undoped and doped zinc oxide nanomaterials as a result of XRD analysis

Sample name	Crystal Size (nm)
Un-doped ZnO NPs	17.50
K-doped ZnO NPs	17.80
Mg-doped ZnO NPs	19.53
Ca-doped ZnO NPs	17.83
Ferula-doped ZnO NPs	17.63
Tagetes-doped ZnO NPs	16.84

EDS analysis revealed the primary components of the synthesized nanomaterials and demonstrated that the elements observed in the spectra were consistent with the precursors. The data obtained revealed a homogeneous distribution of elements on the nanomaterial surface, supporting the expected chemical composition. The presence of the primary elements Zn and oxygen provides evidence of particle purity and of the relationship between morphology and chemical composition (Table 3).

Table 3. Determination of the contents of un-doped and doped nanomaterials according to EDS analysis results

Sample name	Zn (AT %)	O (AT %)	K (AT %)	Mg (AT %)	Ca (AT %)	P (AT %)
Un-doped ZnO NPs	44.06	55.95				
K-doped ZnO NPs	49.42	50.44	0.14			
Mg-doped ZnO NPs	48.34	51.65				
Ca-doped ZnO NPs	40.86	50.09			6.04	3.01
Ferula-doped ZnO NPs	43.78	56.22				
Tagetes-doped ZnO NPs	74.65	25.35				

When all treatment groups were compared with the control group, an increase of 2–22% in the average emergence rate was observed. In particular, the M6 (ZnO NPs100 ppm) and M14 (Mg-doped ZnO NPs 100 ppm) treatment groups had the highest emergence rates, 40% and 39%, respectively, compared with the other treatments and the control group (18%) (Figure 4). Studies have shown that nanopriming treatments accelerate this process by increasing water uptake during the germination and emergence stages (Mahakham et al., 2017) and that different additives used exert differential effects on the seed germination mechanism (Kundu and Bordolui, 2023; Caser et al., 2024). No comparative emergence studies were found in the literature on nanopriming treatments in parsley. In an in vitro germination study, TiO_2 treatment was found to increase the germination rate (Dehkourdi and Mosavi, 2013).

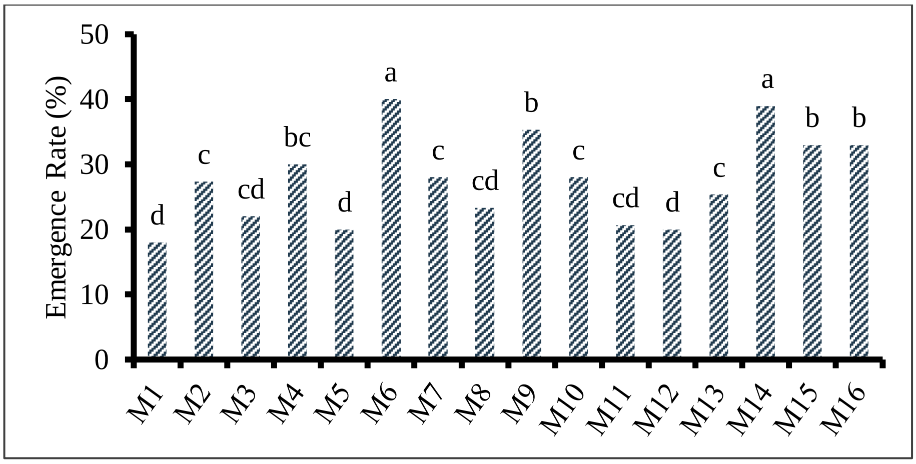

Figure 4. Changes in emergence rate values in parsley after control, hydropriming and nanopriming treatments

It was observed that the treatments could increase seedling length to as much as 10.33 cm (Ferula-doped ZnO NPs at 50 ppm; M9 treatment). An approximately 2 cm increase in root length (Zn 50 ppm treatment, M3) was observed. The effect of the treatments on root system development appears to be important, particularly because a well-developed root system plays an active role in providing water and minerals necessary for stem and leaf development. The increase in seedling length and development ensures the formation of stronger seedlings, thereby increasing photosynthetic capacity and biomass accumulation. Thus, previous studies have emphasized that it is effective in increasing plant growth rate and plant weight in species such as rice and squash (Siddiqui et al., 2014; Mahakham et al., 2017).

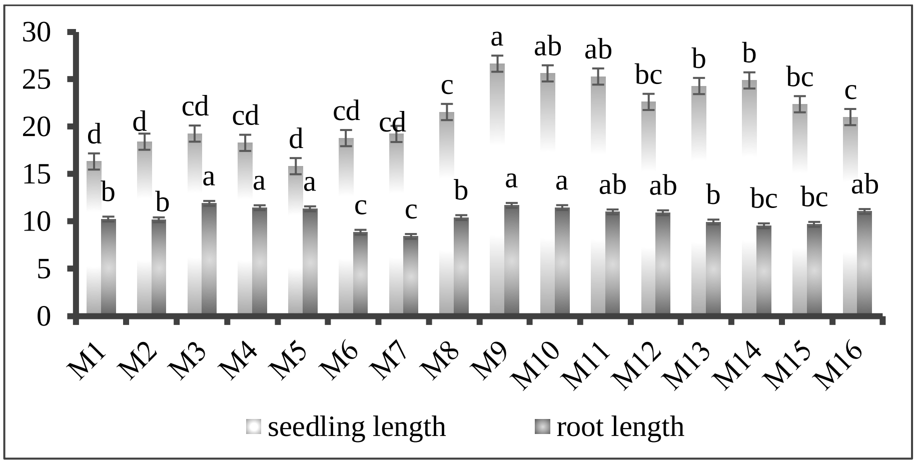

Figure 5. Effect of control, hydropriming and nanopriming treatments on the development of seedling and root length

When the changes in stem and root diameters of the control group following hydropriming and nanopriming treatments were examined, these two traits changed in parallel between the treatments (Figure 6). It was determined that the best results were obtained with the potassium-doped ZnO NPs treatment at 100 ppm (M12) for stem and root diameters. Furthermore, the Ferula-doped ZnO NPs treatment at 100 ppm (M10) and the magnesium-doped ZnO NPs treatment at 50 ppm (M13) showed higher stem and root diameters than those of the other treatments and the control. Greater stem and root diameters are important for plant development, increasing both plant resistance and physiological productivity. Increasing stem diameter increases the capacity of the vascular bundles (xylem and phloem), thereby facilitating material transport. It has been reported that increased root diameter not only increases the uptake of minerals and water from the soil but also increases tolerance to environmental stresses (e.g., flooding and drought) (Taiz et al., 2015; Siddiqui et al., 2014; Kandhol et al., 2022; Nile et al., 2022). However, treatments with titanium oxide nanomaterials have been reported to adversely affect root development in tomato plants at high doses (100 ppm) (Yağız and Çalışkan, 2024). Therefore, it should be considered that the dose and the applied nanoagent may have distinct effects.

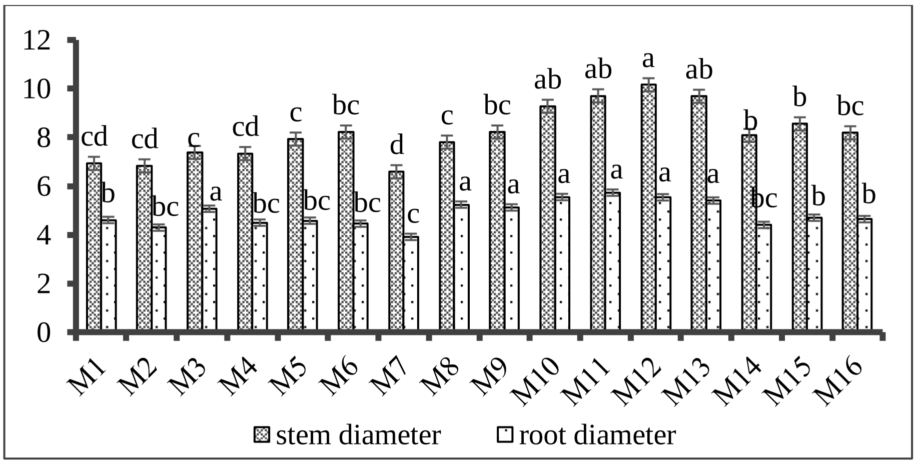

Figure 6. Effect of control, hydropriming and nanopriming treatments on stem and root diameter values

Pre-sowing nanopriming treatments accelerate seedling development by promoting enzyme activation, protein synthesis, and energy production in seeds. This, in turn, alters the fresh and dry weights of seedlings following nanopriming treatments. Since the increase in seedling fresh and dry weights with nanopriming treatments is an indicator of the plant's physiological development as well as its metabolic activity, it is important to examine these characteristics in further studies. The increase in seedling fresh weight after nanopriming treatments indicates enhanced plant water uptake, cell turgor, and overall plant tissue development. Therefore, this increase leads to higher biomass accumulation, metabolic activity, and seedling fresh weight (Siddiqui et al., 2014; Hossain et al., 2015; Tripathi et al., 2016; Mahakham et al., 2017). When the results obtained in this regard were examined (Figure 7), all treatment groups, especially the K-doped ZnO NPs (50 ppm; M11) and Ferula-doped ZnO NPs (50 ppm; M9) treatments, increased seedling fresh and dry weights compared with the control group.

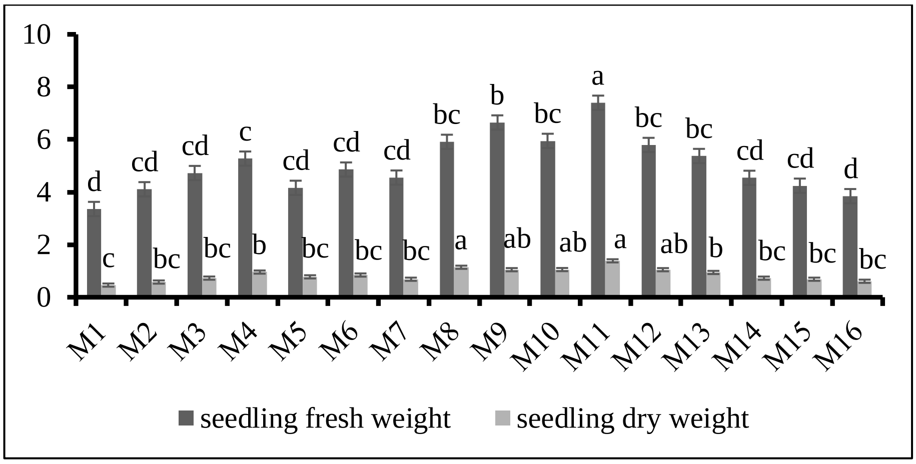

Figure 7. Effect of control, hydropriming and nanopriming treatments on seedling fresh and seedling dry weight changes

Changes in root fresh and dry weights following the control, hydropriming, and nanopriming treatments differed among the treatment groups. The control group (M1) and the 50 ppm Tagetes-doped ZnO NPs treatment (M7) had the lowest root fresh and dry weights (Figure 8). In particular, the 50 ppm potassium-doped ZnO (M11) nanopriming and zinc priming treatments yielded the best results. The Tagetes-doped ZnO NPs treatment at 100 ppm (M8) and the Ferula-doped ZnO NPs (M9 and M10) NPs treatments were also found to yield better results than the other additive groups among the nanopriming treatments. The use of green synthetic organic materials as additives is particularly important in the context of sustainable agriculture and good agricultural practices. Furthermore, using nanomaterials for nanopriming in seed treatment increases metabolic activity in seeds. This, in turn, promotes faster division and elongation of root cells during and after germination. Consequently, root fresh weight increases compared with the control group. The increase in root dry weight is attributable to the effects of nanopriming treatments on carbon accumulation in the roots and on cellulose synthesis in the cell wall (Tripathi et al., 2016; Mahakham et al., 2017; Echeverría-Pérez et al., 2025).

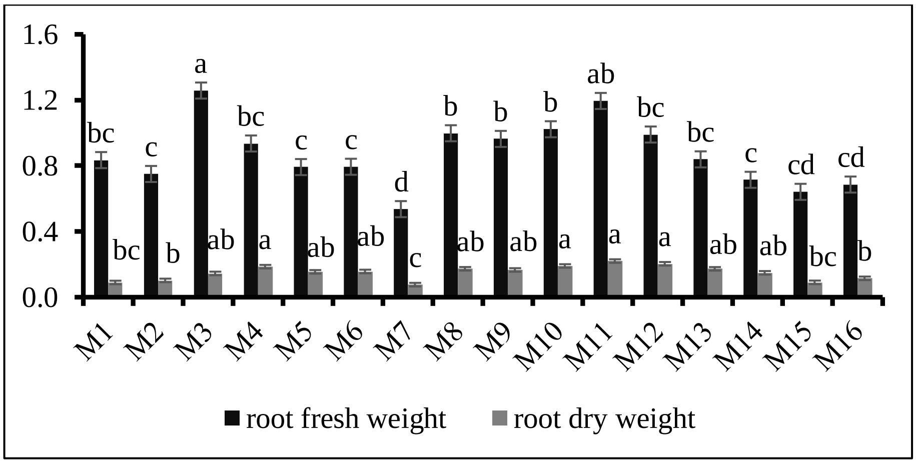

Figure 8. Effect of control, hydropriming and nanopriming treatments on root fresh and root dry weight changes

Nanopriming treatments generally have a stimulatory effect on leaf width, length, and leaf values (Figure 9). This positive effect stems from nanopriming treatments activating physiological and biochemical processes during the seed pre-germination period, increasing energy production, chlorophyll synthesis, and photosynthetic activation, creating a "starting advantage" that carries over into the plant's developmental stages. Thus, nanopriming treatments accelerate leaf development by promoting rapid cell division and cell elongation during seedling development. Studies have shown that the use of nanoparticles, particularly those composed of ZnO, SiO_2, TiO_2, or Fe_3O_4, increases chlorophyll synthesis and photosynthetic efficiency in leaf tissues (Tripathi et al., 2016; Rastogi et al., 2017; Mahakham et al., 2017; Venkatachalam et al., 2017; Song and He, 2021; Adhikary et al., 2022; Ochoa-Chaparro et al., 2025; Ranaware et al., 2025). This allows leaves to have a larger surface area and greater length. In particular, zinc priming and Tagetes- and Ferula-doped zinc nanopriming treatments seem to more accurately reflect this effect. A larger leaf surface area also increases the plant's ability to use light, thereby increasing the rate of photosynthesis (Figure 9). This will directly contribute to biomass production and promote an increase in yield at harvest, as shown in Table 6.

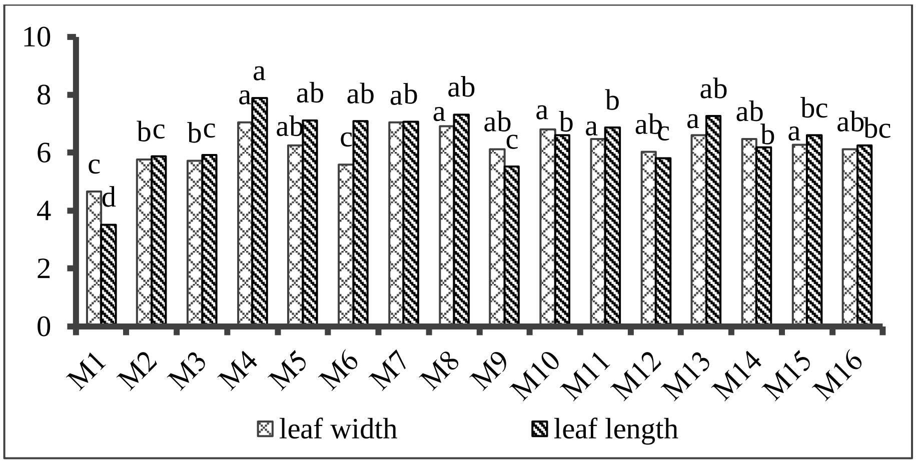

Figure 9. Effect of control, hydropriming and nanopriming treatments on the change of leaf width and length values

Nanopriming treatments lead to the development of a strong root system, increased water and nutrient uptake, and increased photosynthetic efficiency through increased leaf area. This increases plant biomass production and product quality. Nanomaterials enhance metabolic balance by facilitating more efficient transport of micronutrients (e.g., Zn, Fe, Mn) to the leaves, thereby improving yield. Considering that the 50 ppm Ferula-doped ZnO NPs nanopriming treatments increased yield 3.5-fold compared with the control group, the importance of these treatments in plant production, especially in parsley production, is evident. Examination of the effects of other treatments on yield indicated increases ranging from 90.5 to 1093 kg compared with the control group (Table 6).

Table 6. Effect of control, hydropriming and nanopriming treatments on total yield

Treatments	**Total Yield (kg/da)**	**Treatments**	**Total Yield (kg/da)**
Control	969.5 e	Ferula-doped ZnO NPs (50 ppm)	3451.6 a
Hydropriming	1396.3 de	Ferula-doped ZnO NPs (100 ppm)	1655.8 bd
Zn (50 ppm)	1922.5 b	K-doped ZnO NPs (50 ppm)	2062.5 bc
Zn (100 ppm)	1060.0 e	K-doped ZnO NPs (100 ppm)	1501.3 cd
ZnO NPs (50 ppm)	1309.6 de	Mg-doped ZnO NPs (50 ppm)	1485.0 cd
ZnO NPs (100 ppm)	1365.1 de	Mg-doped ZnO NPs (100 ppm)	1839.5 b
Tagetes-doped ZnO NPs (50 ppm)	1455.3 cd	Ca-doped ZnO NPs (50 ppm)	1382.5 de
Tagetes-doped ZnO NPs (100 ppm)	1488.3 cd	Ca-doped ZnO NPs (100 ppm)	1400.5 de

Conclusion

No studies in the reviewed literature on parsley nanopriming reached the yield stage. After the treatments, it was determined that NP treatments supplemented with ferula, potassium, magnesium, and calcium were the most prominent among the NP treatment groups, based on all measurements. In the emergence test, the Ferula- and calcium-supplemented groups had the highest percentage of emergence.

NP treatments supplemented with Ferula were observed to stimulate seedling and root development. Dry matter accumulation was higher in NPs treatments supplemented with potassium.

NPs treatments increased emergence and emergence-related characteristics by 20%, despite the very low emergence of the control group in this study. The effectiveness of the treatments on seedling development and quality varied, particularly with respect to the additive material. While there were no significant differences between treatment doses, the 100 ppm dose yielded better results. In an analysis of per-hectare yield, it was determined that NPs treatments supplemented with ferula and potassium, in particular, increased yield per unit area.

As a result, the increase in plant stem and root diameter resulting from nanopriming treatments provides an advantage that significantly improves the plant's overall growth performance, stress tolerance, and yield potential. Nanopriming treatments increase yield by enhancing metabolic activity, promoting biomass accumulation and cell wall formation. Furthermore, nanopriming treatments enhance the plant's photosynthetic capacity by increasing leaf width and length, thereby supporting nutrient accumulation, energy production, and growth rate. However, future studies should explore various nanomaterials obtained through green synthesis using Ferula and Tagetes in greater detail to understand the specific molecular and biochemical changes they cause in seeds and during plant development.

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Conflict of Interest

There are no conflicts of interest among the authors.

Author Contributions

The authors contributed equally to the conduct and writing of the study.

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