PLANT SCIENCE TODAY
Vol x(x): xx–xx
https://doi.org/10.14719/pst.8145

eISSN 2348-1900

RESEARCH ARTICLE

The effect of manganese oxide and zinc oxide nanoparticles on seed germination characteristics and biochemical changes in *Cichorium intybus* L.

Zahra Ziari, Golnaz Tajadod*, Sedigheh Arbabian & Masoumeh Mirzai

Department of Biology, NT.C., Islamic Azad University, Tehran, Iran

*Correspondence email - tajadodgolnaz@gmail.com

Received: 09 March 2025; Accepted: 25 August 2025; Available online: Version 1.0: 17 October 2025

Cite this article: Zahra Z, Golnaz T, Sedigheh A, Masoumeh M. The effect of manganese oxide and zinc oxide nanoparticles on seed germination characteristics and biochemical changes in *Cichorium intybus* L. Plant Science Today (Early Access). https:/doi.org/10.14719/pst.8145

Abstract

In this experimental study, the effects of two nanoparticles (NPs), zinc oxide and manganese oxide NPs, on the seed germination characteristics, seed development and biochemical changes in chicory (*Cichorium intybus*) were investigated. The concentrations of NPs were set to 0.1, 0.05 and 0.01 g/L. The total number of groups, including the control group, was seven. After preparation and disinfection, the seeds were planted in pots. Subsequently, a series of tests, including microscopic studies, measurement of photosynthetic pigments, quantitative protein assays, enzyme activity assays, carbohydrate assays and germination rate and percentage assays, were conducted. The results show that both NPs (ZnO NPs and MgO NPs), at low concentrations of 0.01 and 0.05 g/L, significantly increased the measured biological factors ($p < 0.05$). The 0.05 g/L dose yielded the most significant increase for both NPs. When the concentration of the NPs was increased to 0.1 g/L, a reduction in the biological factors in chicory was observed. These effects are likely due to the excessive accumulation of nanoparticles, disruption in water absorption and transport and increased generation of free radicals causing toxic stress. These results suggest that the low-dose usage of mentioned NPs led to improved qualitative and quantitative growth of plants, especially chicory.

Keywords: biological factors; chicory; *Cichorium intybus*; manganese oxide nanoparticles; zinc oxide nanoparticles

Introduction

Cichorium intybus L. (common chicory) is a herbaceous, perennial plant that can reach heights of up to half a meter and has blue flowers. It thrives in relatively moist and low-light environments. The roots are brown, sometimes reaching up to one meter in length, with a white interior and a milky sap (1).

Chicory dates to pre-Christian times and has been utilized by various cultures in different regions. Renowned physicians like Galen, Dioscorides and Pliny have historically prescribed it for treating various ailments. Nowadays, numerous individuals in different countries trust in its healing properties and use it traditionally. In the 17th and 18th centuries, chicory gained special attention in Europe for its nutritional properties, even serving as a coffee substitute during periods of high coffee prices. Its cultivation reached its peak in the last two centuries. Although in recent decades, the primary use of chicory cultivation has been for extracting inulin and fructose. In central Europe, its cultivation attracted a huge deal of attention, especially from the food and health industries (2).

Chicory offers numerous health benefits, including liver detoxification, treatment of urinary problems and fever reduction. Additionally, it is approved by the FDA. Chicory contains inulin, which has significant prebiotic properties and plays key roles in improving heart disease, reducing joint pain, constipation, boosting the immune system, relaxation and potentially helping in cancer prevention (3). While traditional medicinal plants like Chichori (*Cichorium intybus*) have long been valued for their mineral content and therapeutic properties, recent advances in nanotechnology have opened new possibilities for enhancing these natural benefits. Nanotechnology has received significant attention due to its ability to produce nanoparticles (NPs) with unique properties in terms of shape, dispersion and chemical composition. These engineered NPs can be combined with plant-derived minerals to create novel formulations with enhanced bioavailability and therapeutic effects. In the following section, we examine how these technological advances can be applied to optimize the delivery and efficacy of minerals traditionally obtained from medicinal plants (4).

Essential minerals in the soil each possess specific physiological properties and are categorized into macronutrients and micronutrients based on their relative concentration required in plant tissues. When the concentration of micronutrients surpasses a certain threshold, it can lead to plant toxicity (5).

Today, NPs enter ecosystems because of result of various industrial activities and are also used as fertilizers in some agricultural practices. Although NPs can positively influence plant growth, it is essential to identify their threshold levels, as exceeding these levels can lead to plant toxicity (4).

Zinc is a crucial element in the formation of diastases. Its deficiency causes disruption in the ribonucleic acid synthesis, leading to disturbances in plant protein formation. Zinc also plays an activating, catalytic and even structural role in plant enzymatic functions. The transport of zinc in plants is complex and its distribution in the phloem is limited (6).

This element plays a role in normal plant metabolism and even reproductive processes and plant growth are affected by its reduction. Zinc deficiency results in diminished photosynthesis, decreased chlorophyll content, structural damage to chlorophyll, reduced chloroplasts in the vascular bundle sheath, decreased membrane protein permeability and excessive accumulation of inorganic phosphorus. Manganese as an essential micro-nutrient for plants and because of its ability to change oxidation states, plays a significant role in oxidation-reduction reactions. It is involved in nitrogen metabolism and mitigates the toxic effects of nitrate by reducing it. Manganese is a cofactor for the enzyme superoxide dismutase, which plays a crucial role in plant antioxidant processes. A deficiency in manganese weakens plants' resilience to environmental stresses (7).

Despite extensive research on nanoparticles in agriculture, studies focusing on their dose-specific effects on medicinal plants like chicory (*Cichorium intybus*) remain limited. This study bridges this gap by systematically evaluating how low vs. high concentrations of ZnO and MnO NPs influence chicory's germination, photosynthetic efficiency and biochemical pathways-a critical step toward optimizing nano-enabled cultivation of high-value medicinal crops. Our work also pioneers the comparison of MnO NPs with ZnO NPs in chicory, offering insights into their distinct roles in enhancing plant resilience and productivity. Given the limited findings on the effects of ZnO NPs and MgO NPs on seed germination, seedling development and biochemical changes in chicory, it is vital to study and research these effects. This experimental study was carried out with these considerations and their significance in mind.

Materials and Methods

This experimental research was conducted using pot cultivation with surface-sterilized seeds of *Cichorium intybus*. During the cultivation phase, the seedlings were foliar treated with aqueous suspensions of ZnO NPs and MgO NPs (purchased from US Nano Research) at three concentrations: 0.1, 0.05 and 0.01 g/L. The nanoparticle solutions were prepared in distilled water. Control groups received distilled water only. After treatment, a series of analyses were performed, including microscopic studies, photosynthetic pigment quantification, protein and enzyme activity assays, carbohydrate content measurements and assessments of germination rate and percentage (7).

Chicory seeds were obtained from Pakan Bazr Esfahan Company. To disinfect the seeds, they were immersed in a 2 % sodium hypochlorite solution and then rinsed three times with distilled water. The treatments were carried out in three replicates. Including the control group, there were seven groups in total. The current research was completely randomized and conducted in three replicates (8).

The disinfected seeds were transferred to pots for planting, with thirty seeds placed in each pot. Each group consisted of three replicates. The pots were kept at a temperature of 25 °C, with a light/dark cycle of 16 hr of light and 8 hr of darkness. The pots were watered three times a week with 30 mL of distilled water and sprayed with the desired concentration of zinc and manganese oxide nanoparticle solutions, as well as Hoagland nutrient solution, every ten days. The solution volume for each pot was one cc. The seeds were carefully examined and evaluated during the germination stage. The samples were maintained and treated until the two-leaf stage. Ultimately, the germination percentage and root length of the seedlings were measured and evaluated. Additionally, a series of tests, like microscopic studies, photosynthetic pigment assays, quantitative protein and enzyme activity assays and carbohydrate assays, were conducted (7).

Quantitative protein and enzyme activity assays

The total protein content and the activity of two enzymes, catalase and peroxidase, were evaluated. Initially, an extract solution was prepared. To create the extract, 1.2 g of Tris, known as a buffering solvent, was mixed with 0.1 g of ascorbic acid, followed by the addition of 17.2 g of sucrose and 0.1 g of cysteine chloride. Then, 26.8 mL of 0.2 normal hydrochloric acid were added. Finally, the solution volume was adjusted to 100 mL with distilled water. Subsequently, 1 g of plant tissue was mixed with 5 mL of the prepared solution and ground in a mortar until completely homogeneous. It was left at room temperature for 10 min. Then, it was centrifuged at 4 °C at a speed of 10000 rpm. The supernatant was stored in a freezer at -20 °C (8).

Peroxidase enzyme activity assay

To measure peroxidase enzyme activity, 0.1 mL of plant extract was mixed with 2 mL of 0.2M acetate buffer, followed by the addition of 0.3 mL of 3 % hydrogen peroxide and 0.1 mL of 0.2M benzidine dissolved in 50 % methanol. The solution was transferred to a test tube, vortexed and allowed to rest at room temperature for 25 min. The absorbance was read at 530 nm using a spectrophotometer (9).

Catalase enzyme activity assay

To measure and evaluate catalase enzyme activity, 0.2 mL of plant extract was mixed with 2.5 mL of phosphate buffer and 0.3 mL of 3 % hydrogen peroxide and transferred to a test tube. After vortexing and resting at room temperature for 25 min, the absorbance was read at 530 nm using a spectrophotometer (10).

Total protein quantification

The Bradford method was used to quantify the total protein. For this purpose, 0.1 mL of extract were mixed with 5 mL of Bradford reagent, transferred to a test tube and vortexed. After a 25-min interval of resting, the absorbance was read at 595 nm using a spectrophotometer (8).

Quantitative measurement of carbohydrates

To measure the carbohydrate content in plants, both soluble and insoluble sugars are assessed. The phenol-sulfuric acid method is employed for sugar quantification. Initially, 1 g of the dried plant material, ground into powder, is mixed with 10 mL of 70 % ethanol. Then, 0.5 mL of the supernatant are taken and diluted to 2 mL with distilled water. Next, 1 mL of 5 % phenol is added, followed by 5 mL of concentrated sulfuric acid after thorough mixing. The solution is allowed to rest for 30 min to dry completely and then the absorbance is read at 485 nm using a spectrophotometer. The final measurement is obtained using a glucose standard curve (10).

For measuring insoluble sugars, the precipitate from the previous step is used. The ethanol solution is filtered and the resulting precipitate is dried and weighed. Next, 10 mL of distilled

water are added and boiled for 15 min in a water bath. The supernatant is then filtered and the volume is adjusted to 25 mL with distilled water. 2 mL of this solution are separated and 1 mL of phenol is introduced. Afterward, 5 mL of concentrated sulfuric acid are added. The mixture is allowed to rest for 30 min at room temperature and then the absorbance is read at 485 nm using a spectrophotometer (8).

Measurement of radicle length and germination percentage

Healthy seeds are used to measure radicle length and germination percentage. After disinfecting the seeds in a sodium hypochlorite solution for 10 min, they are rinsed several times with distilled water. The seeds are then planted in pots under specified conditions and after 3 days, the germination percentage and radicle length are measured. The final germination percentage is calculated using the formula (G = n/N·100%), where (G) is the final germination percentage, (n) is the number of germinated seeds and (N) is the total number of seeds planted (11).

Statistical analysis

All obtained results in this study were statistically analyzed using SPSS software version 21. All reported data were presented as the mean values of experiments ± standard deviation (SD). Subsequently, the Tukey procedure (post hoc analyses) was employed to assess the significant difference among group means ($p < 0.05$) (9).

Results

Measurement of photosynthetic pigments

In these measurements, the levels of chlorophyll a, chlorophyll b and carotenoids were evaluated using fresh plant leaves.

Chlorophyll a measurement results

The addition of both ZnO NPs and MgO NPs significantly improved chlorophyll a level. The maximum increase was observed at a concentration of 0.05 g/L for both nanoparticles, ($p < 0.001$). However, by increasing the concentration of NPs, chlorophyll a level sharply decreased, resulting into a sharp decrease in chlorophyll a level (Fig. 1).

Chlorophyll b measurement results

An increase in chlorophyll b levels was observed with increasing doses of both nanoparticles, especially in MgO NPs. The maximum increase occurred at 0.05 g/L for both nanoparticles ($p < 0.001$). However, higher doses result in a significant decrease in chlorophyll b levels, losing significant difference compared to the control group (Fig. 2).

Carotenoid measurement results

Relative increases in carotenoid levels were observed with the treatment of ZnO NPs and MgO NPs, particularly for ZnO NPs. The maximum increase was at 0.05 g/L, with significant differences at $p < 0.001$ for zinc oxide and $p < 0.01$ for manganese oxide. No statistically significant differences in higher doses were seen (Fig. 3).

Quantitative protein and enzyme activity measurements

Total protein levels and the activities of catalase and peroxidase enzymes were measured in plant extract solutions.

Peroxidase enzyme activity results

Adding more ZnO and MgO NPs made the peroxidase enzyme more active. The enzyme was most active when 0.05 g/L of nanoparticles were used ($p < 0.01$). This performance stopped with high concentration of NPs (Fig. 4).

Catalase enzyme activity results

Catalase activity increased with the treatment of ZnO NPs and MgO NPs, MgO NPs caused a larger increase in catalase activity compared to ZnO NPs. The highest level of catalase activity was observed when using a concentration of 0.05 g/L of both NPs. With significant differences at $p < 0.001$ for manganese oxide and $p < 0.01$ for zinc oxide. This suggests that the higher concentrations of NPs generally led to greater catalase activity (Fig. 5).

Total protein measurement results

Total protein levels enhanced with increasing concentrations of ZnO NPs and MgO NPs up to 0.05 g/L, with significant differences at $p < 0.001$ for manganese oxide and $p < 0.01$ for zinc oxide. A relative decrease in total protein levels was observed at higher doses (Fig. 6).

Quantitative carbohydrate measurements

Soluble and insoluble sugars in the plant were evaluated using the phenol-sulfuric acid method and dry plant material.

Soluble sugar measurement results

The higher doses of ZnO and MgO NPs led to higher soluble sugar levels, with a peak at 0.1 g/L for both types of NPs. Significant differences were observed at $p < 0.001$ for both 0.1 and 0.05 g/L doses (Fig. 7).

Insoluble sugar measurement results

An increasing trend in insoluble sugar levels was observed with increasing doses of both nanoparticles, with a more significant increase for MgO NPs. The highest levels were at 0.1 g/L for both nanoparticles, with significant differences at $p < 0.01$ for zinc oxide and $p < 0.001$ for manganese oxide (Fig. 8).

Root length and germination rate measurements

Healthy seeds were utilized to assess root length and germination rate, with conducted calculations using pot cultures.

Root length measurement results

Pot culture experiments showed a significant increase in root length as the concentration of both NPs increased. The maximum increase was at 0.05 g/L for both nanoparticles ($p < 0.001$). However, further increases in nanoparticle concentration, especially for manganese, caused a decrease in root length growth (Fig. 9).

Germination rate measurement results

In pot culture experiments, a relative and significant increase in germination rate was observed with increasing doses of both nanoparticles. The maximum increase was at 0.05 g/L for both types of nanoparticles, with a significant difference at $p < 0.001$. However, higher doses led to a sharp decline in germination rate for both nanoparticles, with no significant difference noted for manganese oxide (Fig. 10).

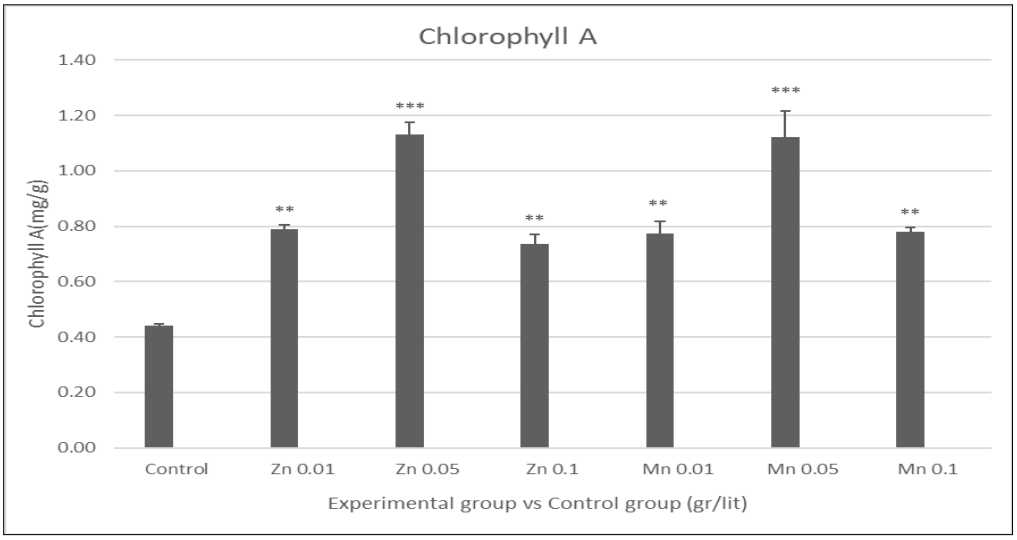

Fig. 1. Results of chlorophyll a measurement. In the two groups of ZnO and MgO NPs at a dose of 0.05 g/L, the highest amount of chlorophyll a was observed ($p < 0.001$). With the increase in the dose of the two nanoparticles, the increasing trend was reversed. *Significant difference with the control group ($p < 0.05$). **Significant difference with the control group ($p < 0.01$). ***Significant difference with the control group ($p < 0.001$).

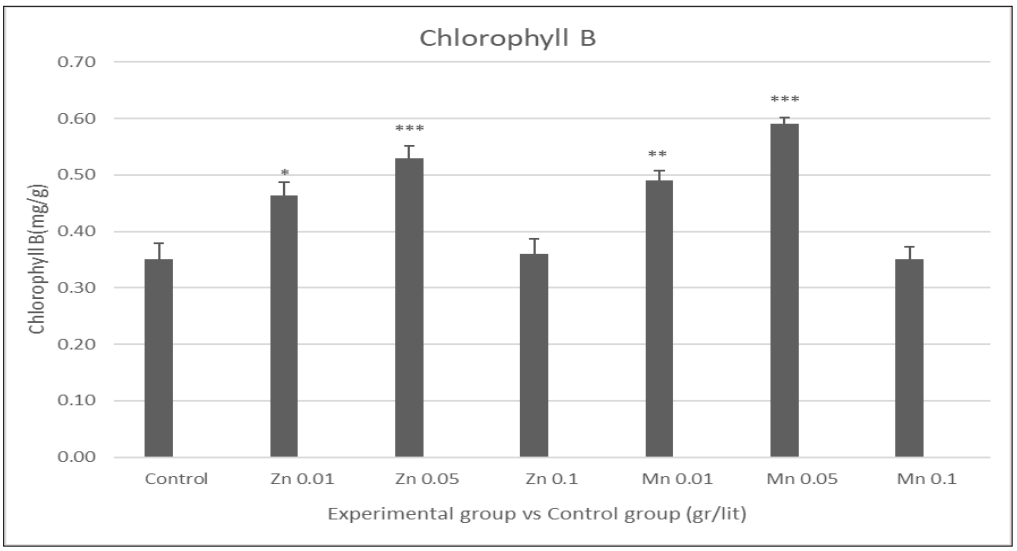

Fig. 2. Results of chlorophyll b measurement. In the two groups of ZnO and MgO NPs at a dose of 0.05 g/L, the highest amount of chlorophyll b was observed ($p < 0.001$). However, with the increase in the dose of the two nanoparticles, the amount of chlorophyll significantly decreased and lost its significant difference with the control group. *Significant difference with the control group ($p < 0.05$). **Significant difference with the control group ($p < 0.01$). ***Significant difference with the control group ($p < 0.001$).

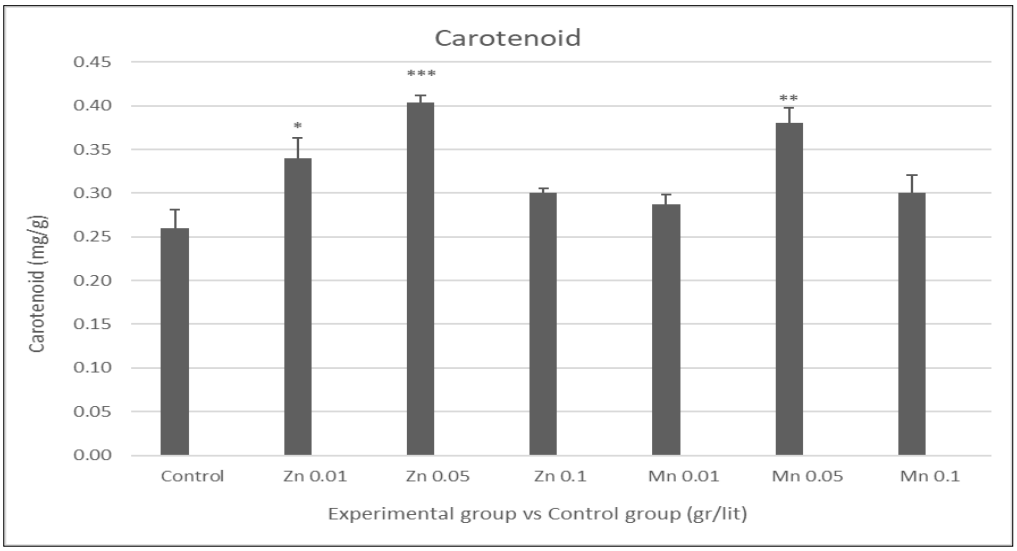

Fig. 3. Results of carotenoid measurement. In the two groups of ZnO and MgO NPs at a dose of 0.05 g/L, the highest amount of carotenoids was observed. With the increase in the dose of the two nanoparticles, the amount of carotenoids decreased and no significant difference was observed. *Significant difference with the control group ($p < 0.05$). **Significant difference with the control group ($p < 0.01$). ***Significant difference with the control group ($p < 0.001$).

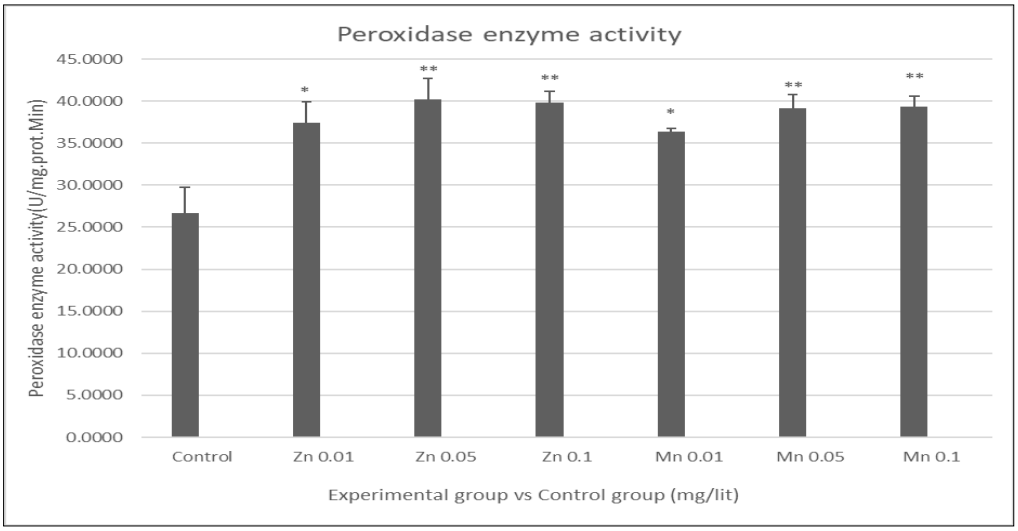

Fig. 4. Results of peroxidase enzyme activity measurement. With the increase in the dose of ZnO and MgO NPs, a relative and significant increase in peroxidase enzyme activity was observed, which was maximum at a dose of 0.05 g/L ($p < 0.01$). However, with further increase in the dose of the two nanoparticles, the enzyme activity did not significantly decrease ($p < 0.01$). *Significant difference with the control group ($p < 0.05$). **Significant difference with the control group ($p < 0.01$). ***Significant difference with the control group ($p < 0.001$).

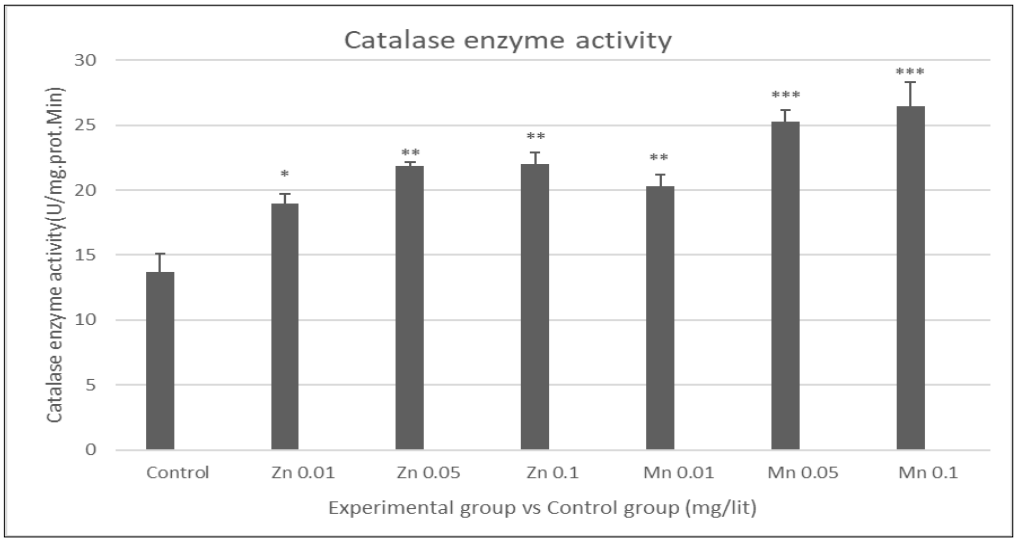

Fig. 5. Results of catalase enzyme activity measurement. In the two groups of ZnO and MgO NPs at a dose of 0.05 g/L, the highest catalase enzyme activity was observed. However, the significant difference for manganese oxide nanoparticles was at the level of $p < 0.001$ and for ZnO NPs at the level of $p < 0.01$. With the increase in the dose of the two nanoparticles, the catalase enzyme activity significantly increased, especially for manganese oxide nanoparticles ($p < 0.001$). *Significant difference with the control group ($p < 0.05$). **Significant difference with the control group ($p < 0.01$). ***Significant difference with the control group ($p < 0.001$).

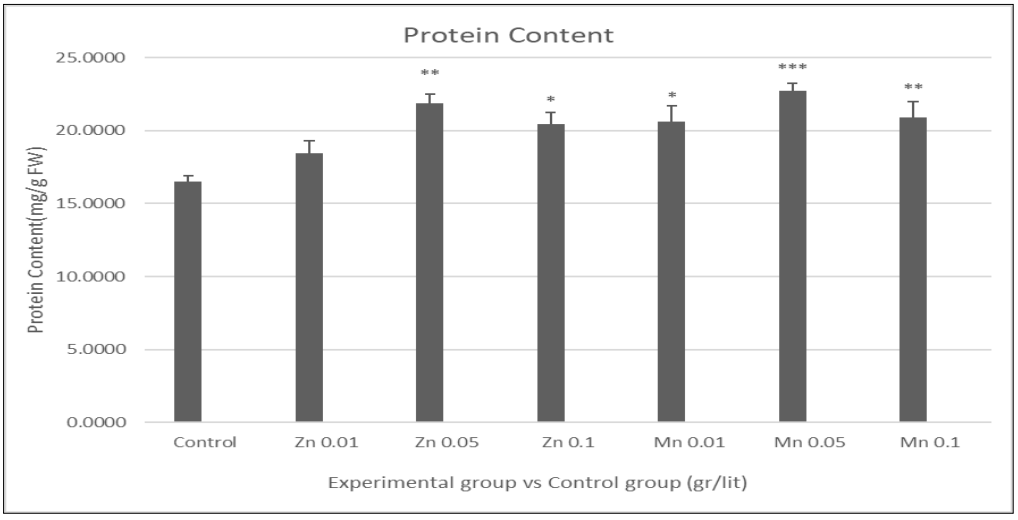

Fig. 6. Results of total protein measurement. In the two groups of ZnO and MgO NPs at a dose of 0.05 g/L, the highest total protein levels were observed. This amount was significant for manganese oxide nanoparticles at the level of $p < 0.001$ and for ZnO NPs at the level of $p < 0.01$. However, treatment with higher doses of both nanoparticles resulted in a downward trend in total protein levels. *Significant difference with the control group ($p < 0.05$). **Significant difference with the control group ($p < 0.01$). ***Significant difference with the control group ($p < 0.001$).

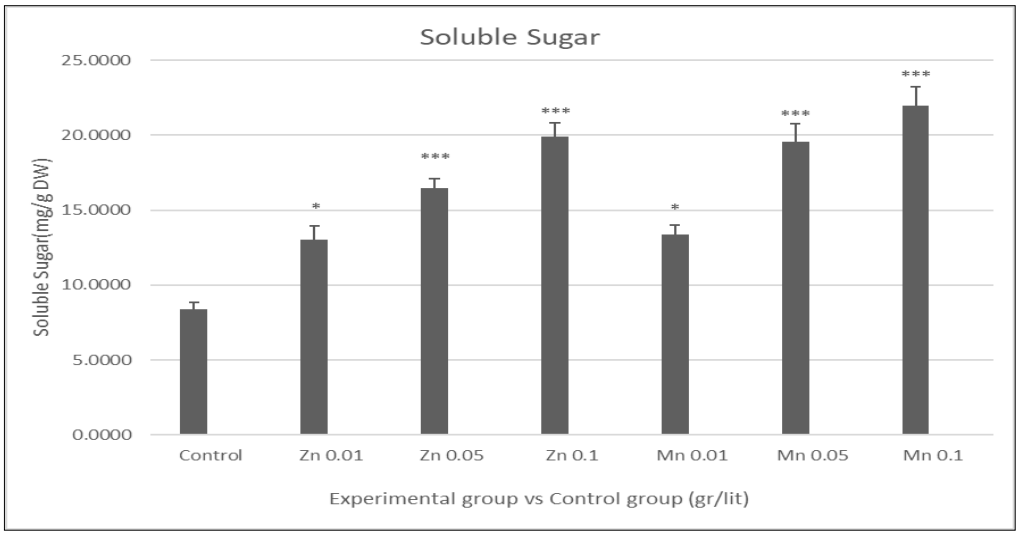

Fig. 7. Results of soluble sugar content measurement. In the two groups of ZnO and MgO NPs at a dose of 0.1 g/L, the highest amount of soluble sugar was observed ($p < 0.001$). Additionally, at a dose of 0.05 g/L, a significant difference was also observed at the level of $p < 0.001$. *Significant difference with the control group ($p < 0.05$). **Significant difference with the control group ($p < 0.01$). ***Significant difference with the control group ($p < 0.001$).

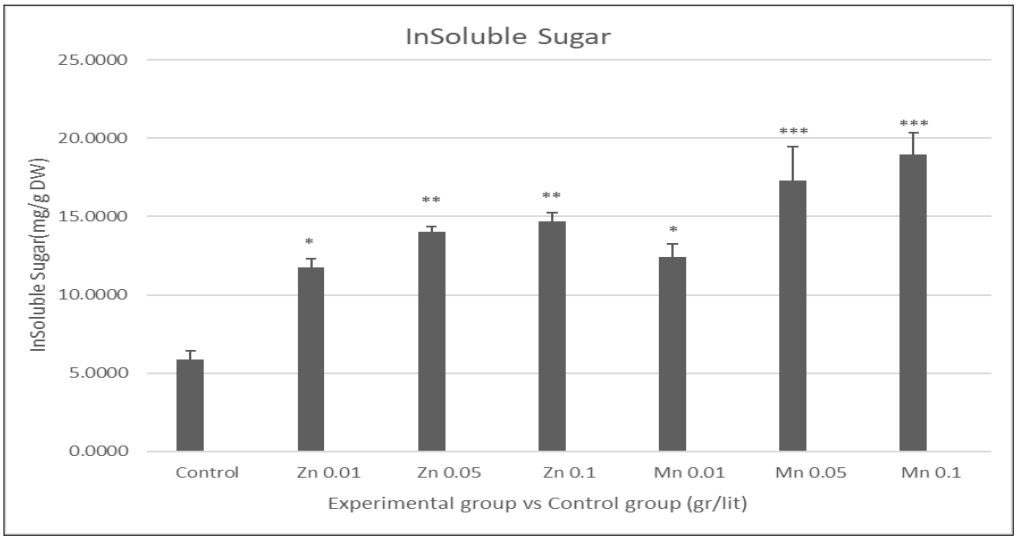

Fig. 8. Results of insoluble sugar content measurement. At a dose of 0.1 g/L, the highest increase in insoluble sugar content was observed. The increase was more significant for manganese oxide nanoparticles ($p < 0.001$). This increasing trend was also observed for ZnO NPs at this dose ($p < 0.01$). *Significant difference with the control group ($p < 0.05$). **Significant difference with the control group ($p < 0.01$). ***Significant difference with the control group ($p < 0.001$).

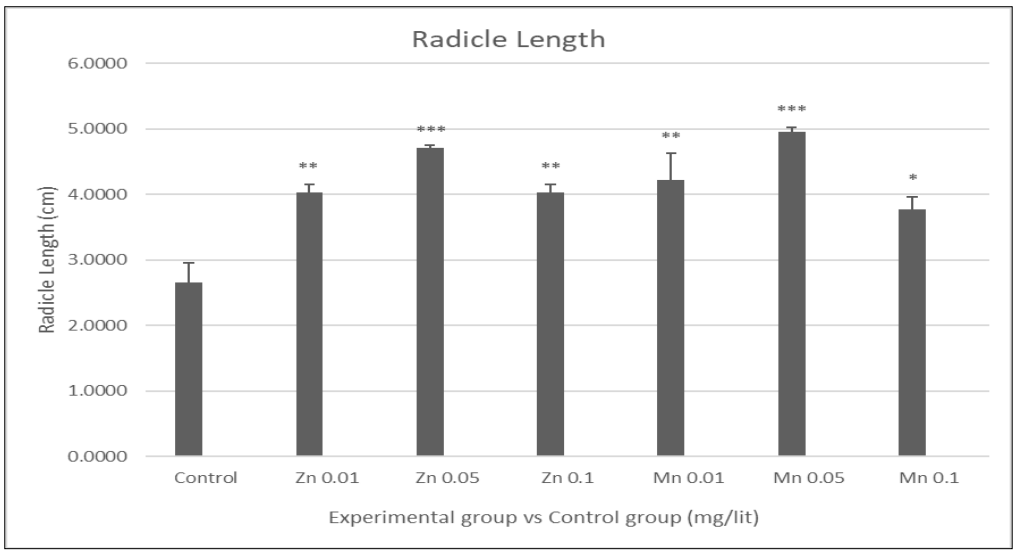

Fig. 9. Results of radicle length measurement. In the two groups of ZnO and MgO NPs at a dose of 0.05 g/L, the highest radicle length growth was observed ($p < 0.001$). With the increase in the dose of the two nanoparticles, the increasing trend was reversed. *Significant difference with the control group ($p < 0.05$). **Significant difference with the control group ($p < 0.01$). ***Significant difference with the control group ($p < 0.001$).

Discussion

In this experimental study, the effects of two nanoparticles, zinc oxide and manganese oxide, on seed germination characteristics, seed development and biochemical changes in chicory plants were investigated. Solutions of ZnO and MgO NPs at concentrations of 0.1, 0.05 and 0.01 g/L were utilized. The number of groups, including the control group, was seven. The experiments were completely randomized and conducted in three replicates.

Measurement of photosynthetic pigments

This study examined the impact of ZnO and MgO nanoparticles on the photosynthetic pigment composition of chicory plants, specifically chlorophyll a, chlorophyll b and carotenoids. Measurements were conducted using fresh leaf samples.

Chlorophyll a level increased significantly following treatment with both nanoparticles, reaching their peak concentration at 0.05 g/L ($p < 0.001$). However, further increases beyond this concentration led to a sharp decline in chlorophyll a, indicating potential nanoparticle-induced stress at higher doses.

Chlorophyll b followed a similar pattern, with a pronounced and statistically significant rise observed at 0.05 g/L ($p < 0.001$). MgO NPs produced a more substantial effect than ZnO NPs. As with chlorophyll a, excessive nanoparticle concentrations caused chlorophyll b levels to decrease, eventually losing statistical significance compared to the control group.

These results suggest that moderate concentrations of ZnO and MgO NPs enhance pigment synthesis, likely supporting photosynthetic performance. However, higher doses may disrupt chlorophyll production, possibly due to oxidative stress or interference with metal ion absorption, underscoring the necessity of dose optimization in nanoparticle application. A study revealed that ZnO NPs positively influenced the levels of photosynthetic pigments in the plant (13).

In a 2021 study observed the positive effects of ZnO NPs on photosynthetic pigments in tomato plants, resulting in enhanced plant growth, increased yield and improved disease resistance (14).

In a 2022 study on maize plants, the application of ZnO NPs via foliar absorption and as a soil fertilizer showed positive effects on the increase of photosynthetic pigments (15).

In a 2021 study on pea plants subjected to salt stress, the beneficial effects of ZnO NPs in mitigating the harmful impacts of soil salinity and enhancing photosynthetic pigments were noted (16).

In a 2022 study on cowpea plants, the effects of MgO NPs applied as foliar spray and soil fertilizer were investigated. It was found that these NPs significantly increase both chlorophyll a and b pigments (17).

In a 2022 study, it was found that MgO NPs not only positively affected the increase in both chlorophyll a and b levels but also reduced the toxic effects of pollutants such as arsenic. This study was conducted on tomato plants (18).

One characteristic of NPs is their high surface area, which improves their tendency to aggregate. This feature can lead to toxicity in plants (19).

Chlorophyll levels are often indicative of environmental stress and plant resistance. Extracellular or intracellular saturation of plants with nanoparticles can disrupt the synthesis of chlorophyll a and b and impair their function. Given the known negative effects of high doses of manganese and ZnO NPs, such an outcome is likely (20).

It has also been proven that high doses of ZnO and MgO NPs cause chlorophyll molecules in pigment-protein complexes of photosystems to disperse, especially during the transcription phase when chlorophyll is photo oxidized. This disruption is caused by interference with the absorption and placement of iron and magnesium ions (21).

Based on the current results and numerous studies, low concentrations of ZnO and MgO NPs increase chlorophyll a and b levels, however at high concentrations, the results are reversed. It is hypothesized that high concentrations of these nanoparticles induce oxidative stress, disrupting the enzymatic processes of photosynthesis (22).

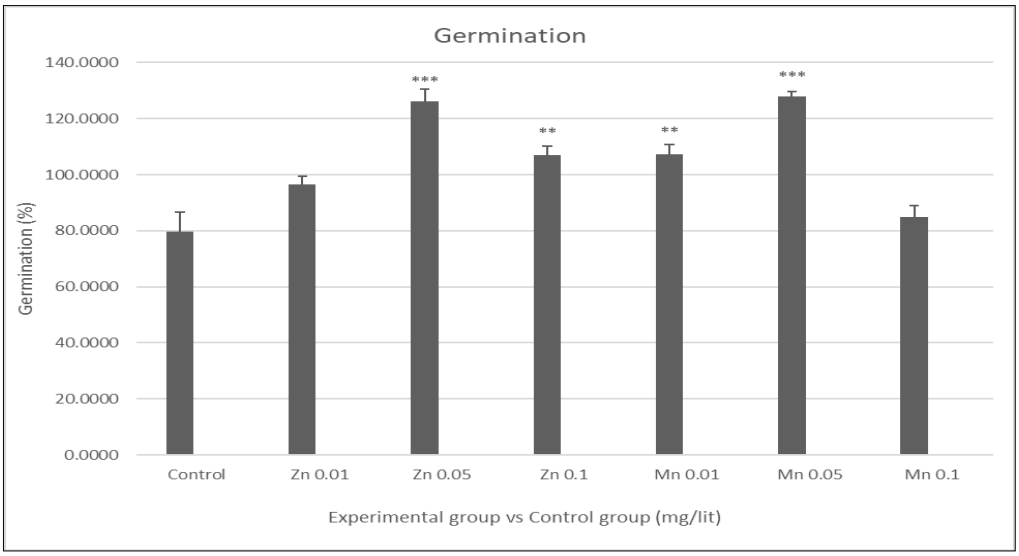

Fig. 10. Results of germination percentage measurement. In the two groups of ZnO and MgO NPs at a dose of 0.05 g/L, the highest germination percentage was observed ($p < 0.001$). With the increase in the dose of the two nanoparticles, the increasing trend was reversed. *Significant difference with the control group ($p < 0.05$). **Significant difference with the control group ($p < 0.01$). ***Significant difference with the control group ($p < 0.001$).

Additionally, at high concentrations of ZnO and MgO NPs, chlorophyll pigments degrade more rapidly. Photosystem II is highly sensitive to environmental stresses and is significantly impacted by the toxic effects of high nanoparticle concentrations, making chlorophyll b more susceptible (23).

Proline is an important amino acid within plant cells, playing a key role in plant protection and defense mechanisms against environmental stresses. Proline also aids in maintaining cell membrane integrity and improves the absorption of nutrients such as potassium and the function of chlorophyll. Research has demonstrated that low doses of ZnO NPs boost this amino acid, but with higher concentrations, its levels and beneficial effects diminish (24).

Elevated photosynthetic products due to increased chlorophyll levels were observed at low concentrations of both nanoparticles. This increase resulted in greater phloem sap production, consequently enlarging the size and number of phloem vessels-particularly with ZnO NPs. However, at high concentrations, destructive effects on chlorophyll reduced photosynthetic output (10).

Based on the mentioned results and previous studies, most research aligns with the present study's findings. Chlorophyll a and b levels increased with the application of these two nanoparticles, but their toxic effects became evident at higher doses.

Quantitative measurement of proteins and enzyme activity

This study assessed the impact of ZnO and MgO nanoparticles on total protein concentration and the enzymatic activity of catalase and peroxidase in chicory plant extracts.

Total protein content exhibited a dose-dependent response to nanoparticle treatment. Both ZnO and MgO NPs promoted protein accumulation up to a concentration of 0.05 g/L, with a statistically significant increase observed for MgO ($p < 0.001$) and ZnO ($p < 0.01$). However, concentrations above this threshold led to a noticeable decline in protein levels, indicating a saturation point or potential nanoparticle-induced stress.

Enzymatic assays showed that peroxidase activity increased with rising nanoparticle doses, reaching peak levels at 0.05 g/L for both types ($p < 0.01$). Further dose escalation slowed this trend, suggesting that high nanoparticle concentrations may hinder enzymatic performance.

Catalase activity followed a similar pattern, with both ZnO and MgO NPs inducing notable enhancement at 0.05 g/L. The effect was more pronounced for MgO NPs ($p < 0.001$), highlighting their stronger influence on antioxidative defense mechanisms. Unlike peroxidase, catalase activity continued to rise at higher nanoparticle concentrations, indicating enzyme-specific responses to oxidative stimuli.

These results suggest that moderate doses of ZnO and MgO NPs can stimulate protein synthesis and antioxidant enzyme activity, reinforcing the plant's biochemical defenses. However, surpassing optimal concentrations may impair these benefits, underlining the need for dosage precision when applying nanoparticles in agricultural systems. In a 2011 study, the effects of ZnO NPs on a group of plants were investigated, confirming their beneficial impact on increasing protein content and enzyme activities (25).

In 2017, research on cotton plants confirmed the positive effects of ZnO NPs on increasing catalase and peroxidase enzyme activities, as well as total protein content (26).

In 2018, the positive effects of ZnO NPs on antioxidant enzyme activities in tomato plants were confirmed, increasing the plant's resistance to environmental stresses (21).

In a 2022 study on a group of plants, MgO NPs increased catalase and peroxidase enzyme activities, enhancing the plant's resistance to oxidative cell damage (27).

Additionally, the effects of MgO NPs on wallflower plants were examined in 2018, revealing an increase in total protein content and antioxidant enzyme activities, although higher doses reduced these effects (28).

Similar results were observed regarding the effects of MgO NPs on the seeds of the plant *Artemisia annua* through foliar spraying and immersion. This research was conducted in 2023. Additionally, forms of manganese nanoparticles, such as manganese sulfate, were examined in this study, which in some cases showed better results compared to MgO NPs (29).

Plants encounter numerous biotic and abiotic stresses throughout their life cycles, leading to various responses by plant cells. The production of reactive oxygen species (ROS) such as H_2O_2 is one of these mechanisms. ROS production occurs within cells and is detoxified by the enzymatic antioxidant system. The removal of toxic amounts and the regulation of appropriate cellular H_2O_2 levels are crucial functions of the enzyme's peroxidase and catalase. Typically, catalase is more significant and functions to convert H_2O_2 into water (23).

According to numerous studies, the toxicity of nanoparticles increases as their size decreases, as smaller nanoparticles possess a higher surface-to-volume ratio (30).

Given the higher solubility of metal oxide nanoparticles, their absorption and toxicity at high doses are also greater, which was evident for ZnO and MgO NPs in the present study. These concentrations of nanoparticles also disrupt the population of beneficial environmental organisms (30).

Reduced growth at high doses may result from water deficiency and the deceleration of processes like mitosis and photosynthesis, as well as increased lipid peroxidation. It has been demonstrated that nanoparticles at high doses increase free radicals, attacking macromolecules such as lipids and increasing lipid peroxidation, which disrupts the nutritional balance and water absorption of the plant. Reduced growth may also stem from decreased protein and chlorophyll levels, as observed in this study at high doses (31).

However, at lower doses, ZnO and MgO NPs increased catalase and peroxidase enzymes, which eliminate hydrogen peroxide in chloroplasts, potentially enhancing chloroplast performance and lifespan. Increased photosynthesis and its products can be secondary results of these effects (32).

Increased protein and chlorophyll may arise from accelerated nitrate uptake and organic nitrogen conversion. Given the structural importance of nitrogen in amino acids, chlorophyll, enzymes and proteins, one can expect quantitative and qualitative increases in many of these products and processes at low doses of these two nanoparticles (8).

Based on the results obtained from the conducted research, low doses of ZnO and MgO NPs increased protein levels and enzymatic activities, but higher doses exhibited toxic and adverse effects, likely due to the mentioned mechanisms.

Antioxidant enzymes such as catalase increase under stress conditions. With the increase in the concentration of manganese oxide and ZnO NPs and the emergence of toxic effects and poisoning caused by these nanoparticles, the levels of antioxidant enzymes like catalase also increased in line with the increase in the doses of these two nanoparticles.

Quantitative measurement of carbohydrates

The carbohydrate content of chicory plants was evaluated by measuring both soluble and insoluble sugars, utilizing the phenol-sulfuric acid method in conjunction with the dry matter of plant organs.

Results showed that soluble sugar levels slightly increased as the concentrations of ZnO and MgO nanoparticles rose. The highest numerical values were recorded at 0.1 g/L for both nanoparticle types. Notably, doses of 0.01 and 0.05 g/L yielded statistically significant increases ($p < 0.001$), suggesting that even lower concentrations can effectively enhance sugar accumulation.

In contrast, insoluble sugar levels exhibited a clear upward trend with increasing nanoparticle concentrations. This effect was more pronounced in treatments involving MgO nanoparticles. At the highest dose of 0.1 g/L, both nanoparticles produced the greatest rise in insoluble sugars, with significance levels of $p < 0.01$ for ZnO and $p < 0.001$ for MgO.

These findings suggest that nanoparticle exposure modulates carbohydrate metabolism in chicory plants, with moderate to high doses promoting sugar accumulation. However, the physiological implications of this metabolic shift—especially in relation to plant stress and energy storage—warrant further investigation. In 2020, a study investigated the effects of ZnO NPs on lettuce seed germination and the quantitative carbohydrate content. The results indicated positive effects of this nanoparticle on carbohydrate reserves in lettuce plants (33).

In 2019, a study on bean plants examined the effects of ZnO NPs on the biological and growth factors of bean plants. The increase in carbohydrate storage was consistent with increasing doses of ZnO NPs (34).

Additionally, in 2020, a study on flax plants and their seeds examined the effects of ZnO NPs on carbohydrate storage. At low doses, the results did not show significant differences, but at higher doses, a significant increase in carbohydrate storage was observed (35).

In a 2018 study, the beneficial effects of MgO NPs on the metabolic reactions of wallflower plants were examined, which consequently led to an increase in the plant's carbohydrate reserves (28).

In 2013, a study on peas and the effects of manganese nanoparticles demonstrated the positive impact of this nanoparticle on enhancing carbohydrate storage, as well as enhancing the plant's resistance to various stresses (36).

As previously noted, environmental stresses result in the production and accumulation of free radicals and reactive oxygen species, which at high concentrations can be harmful to cells, causing lipid peroxidation, enzyme and nucleic acid degradation and even cell membrane damage (37).

The positive effect of two nanoparticles, zinc oxide and manganese oxide, on antioxidant enzymes has been proven, likely due to the increased substrate availability for these enzymes and stronger antioxidant responses. However, with increasing concentrations of these nanoparticles, the amount of hydrogen peroxide increases, revealing their destructive effects (38).

Research indicates, ZnO NPs positively affect auxin biosynthesis, which can also explain the increased growth and performance of plants (19). Zinc also promotes growth regulators and metabolism, influences Krebs cycle reactions through the electron transport chain and plays a crucial role in the division of meristem tissue cells and the increase of hydrocarbon materials. It acts serves as a catalyst for numerous chemical reactions and enzyme systems (39).

Although at low doses, these two nanoparticles, zinc oxide and manganese oxide, enhanced various biological factors in plants, at higher doses, they caused oxidative stress and adverse effects in plants. As previously mentioned, the seed coat and hull play a protective role against toxic substances and environmental pollutants, providing selective absorption. At low concentrations of the nanoparticles examined in this study, it seems that there is no disruption in the absorption of materials and water. However, at high doses, toxic effects of the two nanoparticles become evident, disrupting the absorption of materials and water, which leads to reduced growth of photosynthetic products and biological processes in plants at high concentrations of nanoparticles (40).

In contrast to other biological species, plants have cell walls with selective absorption, but nanoparticles, due to their small size, can more easily and extensively pass through these walls depending on the environmental concentration, leading to the accumulation of these substances at high concentrations. These accumulations at high concentrations result in toxic effects on tissues, cellular organelles and even the cell wall. The transfer of these substances through plasmodesmata may also inflict damage on internal tissues and potentially obstruct channels or pores (41).

At lower doses, these two nanoparticles increased the amount of both types of chlorophyll. This increase enhances the photosynthesis process, which is evident in the plant tissue, especially in the phloem. Even by examining microscopic images, the larger and more numerous phloem vessels at lower doses, especially in ZnO NPs, are noticeable, while at higher doses, the opposite effect was observed and there was no significant difference in manganese oxide nanoparticles.

In the present study, soluble sugars in the plant increased with the concentration of the two nanoparticles. The increase at low doses may be attributed to enhanced processes such as photosynthesis in the plant, but at high concentrations, the accumulation and increased levels of heavy metals for conciseness, the intracellular water balance is disrupted, leading to various changes in the structure of cellular organelles and sugar metabolism enzymes. The reduction in invertase enzyme activity due to the accumulation of heavy metals is one of these

negative effects. As water transfer to different tissues decreases, the accumulation and amount of water-soluble carbohydrates increase, which may serve as an adaptive mechanism to maintain osmotic potential and regulate osmotic pressure under the toxic effects of high concentrations of manganese and ZnO NPs (11).

Due to their small diameter, nanoparticles exhibit greater impact and transfer efficiency compared to larger particles. At low concentrations, nanoparticles such as manganese oxide (MnO) and zinc oxide (ZnO) enhance photosynthetic activity. Their stimulating effect arises from their ability to enter chloroplasts-particularly photosystem II-where they facilitate electron capture and transfer, thereby improving photosynthetic efficiency. However, at elevated concentrations, these nanoparticles can cause cellular damage and even plant mortality. Manganese plays a critical role in the structure and function of over 35 plant enzymes, explaining why low concentrations of MnO nanoparticles enhance various biological processes. This element's involvement in metabolic pathways also improves both functional and morphological traits in plants. Furthermore, the increased activity of antioxidant enzymes at optimal nanoparticle concentrations helps mitigate oxidative damage caused by free radicals. It is important to note that these beneficial effects occur only at low concentrations; at higher doses, the nanoparticles' positive influences are reversed, leading to toxicity.

Measurement of radicle length and germination percentage

The study revealed a concentration-dependent effect of ZnO and MgO nanoparticles on radicle length and germination percentage in chicory seeds. At a dose of 0.05 g/L, both nanoparticles significantly enhanced radicle elongation ($p < 0.001$), indicating an optimal level for promoting early root development. However, further increases beyond this concentration led to a substantial decline in radicle growth, suggesting potential phytotoxic effects at higher nanoparticle levels.

A similar trend was observed for germination percentage. The highest germination rate occurred at 0.05 g/L for both types of nanoparticles, with a statistically significant improvement compared to the control ($p < 0.001$). Nonetheless, exceeding this concentration caused a marked reduction in germination percentage. Specifically, at elevated levels of manganese oxide nanoparticles, the difference in germination rate compared to the control group was no longer significant, underscoring the negative impact of excessive nanoparticle exposure.

These findings align with existing literature on nanoparticle -plant interactions, where moderate concentrations can act as growth stimulants, while excessive doses disrupt normal physiological functions. The balance between enhancement and toxicity emphasizes the importance of precise dose management in nanoparticle applications for agricultural purposes. In 2016, a study investigated the effects of ZnO NPs on the radicle growth of wheat seeds. Positive effects were observed at low doses, but toxic effects of ZnO NPs were identified at high doses (42).

In 2016, the effects of manganese oxide and ZnO NPs on lettuce seeds were studied. The findings showed positive effects of these two nanoparticles on radicle growth and germination percentage, although manganese oxide nanoparticles yielded better results. However, both nanoparticles exhibited toxic effects at high doses (43).

In 2019, positive effects of ZnO NPs on biological factors of carrots, including radicle growth and germination percentage, were observed and confirmed. This increase was consistent with the increase in nanoparticle dose (44).

In 2020, a study explored the effects of a group of nanoparticles, including MgO and ZnO NPs, on the biological factors and growth of two plants, mung bean and lentil. The root length and germination percentage in both plants and for both nanoparticles explained a significant increase (45).

The varied responses of different plants to various concentrations of metals and their oxides in nano form are related to the permeability of the seed coat and its interaction with different compounds. The seed coat plays an important role in protecting the plant embryo. A characteristic that also applies to nanoparticles. In the parenchyma of the seed coat, there are intracellular spaces smaller than 10 µm in size, which, when surrounded by an aqueous solution, facilitate various penetration processes of nano-compounds (46).

Research has indicated that nanoparticles can penetrate the cell wall and reach the DNA of plant cells. Some nanoparticles have been proven to penetrate the plasma membrane and enter the nucleus. The aforementioned research confirmed that the rate at which nanoparticles enter the seed coat varies and can influence germination depending on this entry rate. This variability can account for the different properties of ZnO NPs and MgO NPs at different concentrations and their impact on germination and plant growth. Studies have demonstrated that NPs are capable of passing through the membrane of plasma and entering the nucleus (47).

Since roots are the initial parts of the plant to receive nanoparticles from the environment, their reaction can significantly determine the positive or negative effects of nanoparticles and act as a reliable indicator for their evaluation. The presence of toxic effects at high concentrations of these two nanoparticles diminishes the plant's water uptake process and inhibits the longitudinal growth of plant organs (48).

High concentrations of zinc and manganese nanoparticles negatively impacted on the root growth, likely due to the toxic effects of these nanoparticles at high doses. Because of the presence of mucilage secreted from the root tip and root hairs and the presence of hydrated pectic and polysaccharide substances, enhance the absorption of NPs in these regions, increasing the likelihood of adverse effects (19).

Nanoparticles usually pass through the apoplastic pathways from the cortex and epidermis of the root but must pass through the protoplast of endodermal cells to reach more central regions such as the vascular tissues. Tissue section examinations revealed that as NPs doses enhanced, tissue damage and destruction of various parts of the root and stem occurred, resulting in reduced root growth, germination and many plants metabolic processes. These toxic effects were not observed at lower doses. The toxic effects of high doses of these two nanoparticles are linked to the destruction and vacuolation of cellular parenchyma. Typically, ZnO and MgO NPs create holes in the cell wall, increasing permeability and after entering the cell, they are transported through plasmodesmata (49).

By evaluating characteristics such as root growth or germination percentage, the positive or negative properties of compounds added to the plant can be quickly and easily assessed. Essentially, germination is a complex biological process that begins with water absorption by the plant seeds and reaches its maximum with the emergence of the root. The seed coat is one of the factors that can influence the passage of various substances, including nanoparticles. Therefore, nanoparticles usually do not have a noticeable effect on root growth and germination percentage at very low concentrations. On the other hand, at very high doses, they are likely to aggregate, leading to toxic effects. This clustering may even occur within cellular organelles such as vesicles (38).

According to findings of the current research, which were consistent with most research, low doses of ZnO and MgO NPs enhance root length and germination percentage, but at high doses, it is believed that the mechanisms mentioned lead to a decrease in root length and germination percentage.

Conclusion

This study highlights the concentration-dependent effects of zinc oxide (ZnO) and magnesium oxide (MgO) nanoparticles on the photosynthetic pigment profile of chicory plants. Moderate doses, particularly 0.05 g/L, significantly enhanced levels of chlorophyll a and b, as well as carotenoids, suggesting improved photosynthetic efficiency and potential growth stimulation. MgO nanoparticles demonstrated a slightly stronger effect on chlorophyll b compared to ZnO. However, at higher concentrations, pigment levels declined sharply-indicating that excessive nanoparticle exposure may induce oxidative stress or interfere with key metabolic pathways such as metal ion uptake and pigment synthesis. These outcomes emphasize the importance of precise dosing strategies when employing nanoparticles in agricultural systems to balance their stimulatory effects with potential phytotoxicity. Overall, the findings support the potential of ZnO and MgO NPs as biostimulants at optimized concentrations, while cautioning against their overuse.

Authors' contributions

ZZ conducted all the main experimental work, performed data analysis and drafted the manuscript. The original idea for the study was also conceived by ZZ. GT supervised the experimental methods, provided guidance and oversaw the research process. SA and MM assisted in conducting the experiments and provided additional guidance throughout the study. All authors read and approved the final manuscript.

Compliance with ethical standards

Conflict of interest: Authors do not have any conflict of interest to declare.

Ethical issues: None

References

1. Janda K, Gutowska I, Geszke-Moritz M, Jakubczyk K. The common cichory (*Cichorium intybus* L.) as a source of extracts with health-promoting properties—A review. Molecules. 2021;26(6):1814. https://doi.org/10.3390/molecules26061814
2. Al-Snafi AE. Medical importance of *Cichorium intybus* – A review. IOSR J Pharm. 2016;6(3):41-56. https://doi.org/10.9790/3013-0702014358
3. Street RA, Sidana J, Prinsloo G. *Cichorium intybus*: traditional uses, phytochemistry, pharmacology and toxicology. Evid Based Complement Alternat Med. 2013;2013:579319. https://doi.org/10.1155/2013/579319
4. McNeil SE. Nanotechnology for the biologist. J Leukoc Biol. 2005;78 (3):585-94. https://doi.org/10.1189/jlb.0205074
5. Mukhopadhyay SS. Nanotechnology in agriculture: prospects and constraints. Nanotechnol Sci Appl. 2014;7:63-71. https://doi.org/10.2147/NSA.S39409
6. Sabir S, Arshad M, Chaudhari SK. Zinc oxide nanoparticles for revolutionizing agriculture: synthesis and applications. Sci World J. 2014;2014:925494. https://doi.org/10.1155/2014/925494
7. Rashed MH, Hoque TS, Jahangir MMR, Hashem MA. Manganese as a micronutrient in agriculture: crop requirement and management. J Environ Sci Nat Resour. 2019;12(1-2):225-42. https://doi.org/10.3329/jesnr.v12i1-2.52040
8. Nemček L, Šebesta M, Urík M, Bujdoš M, Dobročka E, Vávra I. Impact of bulk ZnO, ZnO nanoparticles and dissolved Zn on early growth stages of barley—A pot experiment. Plants. 2020;9(10):1365. https://doi.org/10.3390/plants9101365
9. Alavi E, Tajadod G, Jafari Marandi S, Arbabian S. *Vicia faba* seed: a bioindicator of phytotoxicity, genotoxicity and cytotoxicity of light crude oil. Environ Sci Pollut Res. 2023;30(8):21043-51. https://doi.org/10.1007/s11356-022-22456-1
10. Tajadod G, Farzamisepehr M, Kalami Z. B-Glucan contents in calli of *Oryza sativa* L.var Hashemi under different nutritional treatments. Iran J Plant Physiol. 2012;2(3):471-5.
11. Jafari Marandi S, Tajadod G, Peyvandi M. Development of male and female reproduction organs in Moldavian dragonhead, *Dracocephalum moldavica* L. (Lamiaceae). Iran J Plant Biol. 2023;14(4):21-38.
12. Roohizadeh G, Arbabian S, Tajadod G, Majd A, Salimpour F. The study of nano silica effects on the total protein content and the activities of catalase, peroxidase and superoxide dismutase of *Vicia faba* L. Tropical Plant Res. 2015;2(1):47-50.
13. Rai-Kalal P, Jajoo A. Priming with zinc oxide nanoparticles improve germination and photosynthetic performance in wheat. Plant Physiol Biochem. 2021;160:341-51. https://doi.org/10.1016/j.plaphy.2021.01.034
14. Parveen A, Siddiqui ZA. Zinc oxide nanoparticles affect growth, photosynthetic pigments, proline content and bacterial and fungal diseases of tomato. Arch Phytopathol Plant Prot. 2021;54(17-18):1519-38. https://doi.org/10.1080/03235408.2021.1920935
15. Azam M, Bhatti HN, Khan A, Zafar L, Iqbal M. Zinc oxide nano-fertilizer application (foliar and soil) effect on the growth, photosynthetic pigments and antioxidant system of maize cultivar. Biocatal Agric Biotechnol. 2022;42:102343. https://doi.org/10.1016/j.bcab.2022.102343
16. Elshoky HA, Yotsova E, Farghali MA, Farroh KY, El-Sayed K, Elzorkany HE, et al. Impact of foliar spray of zinc oxide nanoparticles on the photosynthesis of *Pisum sativum* L. under salt stress. Plant Physiol Biochem. 2021;167:607-18. https://doi.org/10.1016/j.plaphy.2021.08.021
17. Samsoon S, Azam M, Khan A, Ashraf M, Bhatti HN, Alshawwa SZ, et al. Green-synthesized MnO_2 nanofertilizer impact on growth, photosynthetic pigment and non-enzymatic antioxidant of *Vigna unguiculata* cultivar. Biomass Convers Biorefinery. 2024;14:26943-52. https://doi.org/10.1007/s13399-022-03686-5

18. Faizan M, Bhat JA, El-Serehy HA, Moustakas M, Ahmad P. Magnesium oxide nanoparticles (MgO-NPs) alleviate arsenic toxicity in soybean by modulating photosynthetic function, nutrient uptake and antioxidant potential. Metals. 2022;12(12):2030. https://doi.org/10.3390/met12122030

19. Jiang M, Wang J, Rui M, Yang L, Shen J, Chu H, et al. OsFTIP7 determines metallic oxide nanoparticles response and tolerance by regulating auxin biosynthesis in rice. J Hazard Mater. 2021;403:123946. https://doi.org/10.1016/j.jhazmat.2020.123946

20. Xu QS, Hu JZ, Xie KB, Yang HY, Du KH, Shi GX. Accumulation and acute toxicity of silver in *Potamogeton crispus* L. J Hazard Mater. 2010;173(1-3):186-93. https://doi.org/10.1016/j.jhazmat.2009.08.073

21. Wang XP, Li QQ, Pei ZM, Wang SC. Effects of zinc oxide nanoparticles on the growth, photosynthetic traits and antioxidative enzymes in tomato plants. Biol Plant. 2018;62:801-8. https://doi.org/10.1007/s10535-018-0804-5

22. Esparham E, Saeidisar S, Mahmoodzadeh H, Hadi MR. The effects of zinc oxide (ZnO) nanoparticles on the germination, biochemical and ultrastructural cell characteristics of *Ricinus communis*. Cell Tissue J. 2017;8(2):151-64.

23. Mauchamp A, Methy M. Submergence-induced damage of photosynthetic apparatus in *Phragmites australis*. Environ Exp Bot. 2004;51(3):227-35. https://doi.org/10.1016/j.envexpbot.2003.11.002

24. Khan MA, Shirazi MU, Khan MA, Mujtaba SM, Islam E, Mumtaz S, et al. Role of proline, K/Na ratio and chlorophyll content in salt tolerance of wheat (*Triticum aestivum* L.). Pak J Bot. 2009;41(2):633-8.

25. Ansari SA, Husain Q, Qayyum S, Azam A. Designing and surface modification of zinc oxide nanoparticles for biomedical applications. Food Chem Toxicol. 2011;49(9):2107-15. https://doi.org/10.1016/j.fct.2011.05.029

26. Venkatachalam P, Priyanka N, Manikandan K, Ganeshbabu I, Indiraarulselvi P, Geetha N, et al. Enhanced plant growth promoting role of phycomolecules coated zinc oxide nanoparticles with P supplementation in cotton (*Gossypium hirsutum* L.). Plant Physiol Biochem. 2017;110:118-27. https://doi.org/10.1016/j.plaphy.2016.07.020

27. Zhang Y, Chen L, Sun R, Lv R, Du T, Li Y, et al. Multienzymatic antioxidant activity of manganese-based nanoparticles for protection against oxidative cell damage. ACS Biomater Sci Eng. 2022;8(2):638-48. https://doi.org/10.1021/acsbiomaterials.1c01123

28. Tian H, Ghorbanpour M, Kariman K. Manganese oxide nanoparticle-induced changes in growth, redox reactions and elicitation of antioxidant metabolites in deadly nightshade (*Atropa belladonna* L.). Ind Crops Prod. 2018;126:403-14. https://doi.org/10.1016/j.indcrop.2018.10.021

29. Salehi H, Cheheregani Rad A, Raza A, Djalovic I, Prasad PV. The comparative effects of manganese nanoparticles and their counterparts (bulk and ionic) in *Artemisia annua* plants via seed priming and foliar application. Front Plant Sci. 2023;13:1098772. https://doi.org/10.3389/fpls.2022.1098772

30. Yang L, Watts DJ. Particle surface characteristics may play an important role in phytotoxicity of alumina nanoparticles. Toxicol Lett. 2005;158(2):122-32. https://doi.org/10.1016/j.toxlet.2005.03.003

31. Oberdörster G, Stone V, Donaldson K. Toxicology of nanoparticles: a historical perspective. Nanotoxicology. 2007;1(1):2-25. https://doi.org/10.1080/17435390701314761

32. Lei Z, Mingyu S, Xiao W, Chao L, Chunxiang Q, Liang C, et al. Antioxidant stress is promoted by nano-anatase in spinach chloroplasts under UV-B radiation. Biol Trace Elem Res. 2008;121:69-79. https://doi.org/10.1007/s12011-007-8021-7

33. Rawashdeh RY, Harb AM, AlHasan AM. Biological interaction levels of zinc oxide nanoparticles: lettuce seeds as case study. Heliyon. 2020;6(5):e03924. https://doi.org/10.1016/j.heliyon.2020.e03924

34. Salama DM, Osman SA, Abd El-Aziz ME, Abd Elwahed MS, Shaaban EA. Effect of zinc oxide nanoparticles on the growth, genomic DNA, production and the quality of common dry bean (*Phaseolus vulgaris*). Biocatal Agric Biotechnol. 2019;18:101083. https://doi.org/10.1016/j.bcab.2019.101083

35. Zaeem A, Drouet S, Anjum S, Khurshid R, Younas M, Blondeau JP, et al. Effects of biogenic zinc oxide nanoparticles on growth and oxidative stress response in flax seedlings vs. *in vitro* cultures: a comparative analysis. Biomolecules. 2020;10(6):918. https://doi.org/10.3390/biom10060918

36. Pradhan S, Patra P, Das S, Chandra S, Mitra S, Dey KK, et al. Photochemical modulation of biosafe manganese nanoparticles on Vigna radiata: A detailed molecular, biochemical and biophysical study. Environ Sci Technol. 2013;47(22):13122-31. https://doi.org/10.1021/es403146b

37. Del Rio LA, Sandalio LM, Corpas FJ, Palma JM, Barroso JB. Reactive oxygen species and reactive nitrogen species in peroxisomes production, scavenging and role in cell signaling. Plant Physiol. 2006;141(2):330-5. https://doi.org/10.1104/pp.106.078204

38. Corredor E, Testillano PS, Coronado MJ, González-Melendi P, Fernández-Pacheco R, Marquina C, et al. Nanoparticle penetration and transport in living pumpkin plants: in situ subcellular identification. BMC Plant Biol. 2009;9(1):45. https://doi.org/10.1186/1471-2229-9-45

39. Heidarian AR, Kord H, Mostafavi KH, Lak AP. Investigating Fe and Zn foliar application on yield and its components of soybean (*Glycine max* L.) at different growth stages. J Agric Biotechnol Sustain Dev. 2011;3:189-97.

40. Zareii A, Abbaspour H, Peyvandi M, Majd A. Oxidative stress responses and toxicity of green synthesized silver nanoparticles (AgNPs) on basil (*Ocimum basilicum*) seedlings. J Chem Health Risks. 2023;13(4):691-9. https://doi.org/10.22034/jchr.2023.1973089.1574

41. Rico CM, Majumdar S, Duarte-Gardea M, Peralta-Videa JR, Gardea-Torresdey JL. Interaction of nanoparticles with edible plants and their possible implications in the food chain. J Agric Food Chem. 2011;59(8):3485-98. https://doi.org/10.1021/jf104517j

42. Prakash MG, Chung IM. Determination of zinc oxide nanoparticles toxicity in root growth in wheat (*Triticum aestivum* L.) seedlings. Acta Biol Hung. 2016;67(3):286-96. https://doi.org/10.1556/018.67.2016.3.7

43. Liu R, Zhang H, Lal R. Effects of stabilized nanoparticles of copper, zinc, manganese and iron oxides in low concentrations on lettuce (*Lactuca sativa*) seed germination: nanotoxicants or nanonutrients? Water Air Soil Pollut. 2016;227:1-14. https://doi.org/10.1007/s11270-015-2765-7

44. Siddiqui ZA, Parveen A, Ahmad L, Hashem A. Effects of graphene oxide and zinc oxide nanoparticles on growth, chlorophyll, carotenoids, proline contents and diseases of carrot. Sci Hortic. 2019;249:374-82. https://doi.org/10.1016/j.scienta.2019.01.059

45. Rani P, Kaur G, Rao KV, Singh J, Rawat M. Impact of green synthesized metal oxide nanoparticles on seed germination and seedling growth of *Vigna radiata* (mung bean) and *Cajanus cajan* (red gram). J Inorg Organomet Polym Mater. 2020;30:4053-62. https://doi.org/10.1007/s10904-020-01519-8

46. Van Dongen JT, Ammerlaan AM, Wouterlood M, Van Aelst AC, Borstlap AC. Structure of the developing pea seed coat and the post-phloem transport pathway of nutrients. Ann Bot. 2003;91(6):729-37. https://doi.org/10.1093/aob/mcg078

47. Slowing I, Trewyn BG, Lin VSY. Effect of surface functionalization of MCM-41-type mesoporous silica nanoparticles on the endocytosis by human cancer cells. J Am Chem Soc. 2006;128(46):14792-3. https://doi.org/10.1021/ja065209p

48. Morla S, Rao CR, Chakrapani R. Factors affecting seed germination and seedling growth of tomato plants cultured *in vitro* conditions. J Chem Biol Phys Sci. 2011;1(2):328.

49. Lin D, Xing B. Root uptake and phytotoxicity of ZnO nanoparticles. Environ Sci Technol. 2008;42(15):5580-5. https://doi.org/10.1021/es800422x

Additional information

Peer review: Publisher thanks Sectional Editor and the other anonymous reviewers for their contribution to the peer review of this work.

Reprints & permissions information is available at https://horizonepublishing.com/journals/index.php/PST/open_access_policy

Publisher's Note: Horizon e-Publishing Group remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

Indexing: Plant Science Today, published by Horizon e-Publishing Group, is covered by Scopus, Web of Science, BIOSIS Previews, Clarivate Analytics, NAAS, UGC Care, etc

See https://horizonepublishing.com/journals/index.php/PST/indexing_abstracting

Publisher information: Plant Science Today is published by HORIZON e-Publishing Group with support from Empirion Publishers Private Limited, Thiruvananthapuram, India.