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Original Research Article

Effects of Iron Nanoparticles (FeNPs) on the Growth and Biochemical Changes of Maize (*Zea Mays)* Seedlings

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Abstract

Iron nanoparticles (FeNPs) have emerged as promising nanomaterials for enhancing plant growth and productivity. However, their effects on the growth and biochemical responses of maize (*Zea mays L.*) seedlings remain insufficiently understood. This study investigated the effects of FeNPs on the growth and biochemical responses of three maize varieties. Iron nanoparticles were synthesized from ferric chloride ($FeCl_3$), ferrous sulfate heptahydrate ($FeSO_4 \cdot 7H_2O$), and ammonium hydroxide (NH_4OH) at the Department of Pure and Industrial Chemistry, Bayero University, Kano. Three maize varieties (ZAMMAS27, ZAMMAS51, and a local variety) were evaluated under four treatments comprising a control (0 mg/L) and FeNP concentrations of 50, 75, and 100 mg/L using a randomized complete block design (RCBD) with three replicates. Growth and biochemical parameters were assessed 20 days after sowing. Superoxide dismutase (SOD) activity was determined using a commercial assay kit at Ahmadu Bello University, Zaria. Data were analyzed using analysis of variance (ANOVA), and treatment means were separated using the least significant difference (LSD) test at $P < 0.05$. The results showed that germination percentage declined with increasing FeNP concentration, whereas shoot fresh weight and SOD activity generally increased at higher concentrations. Chlorophyll content decreased with increasing FeNP concentration, indicating a concentration-dependent response. Among the tested genotypes, ZAMMAS51 exhibited the highest SOD activity at 100 mg/L FeNPs. These findings demonstrate that FeNPs exert concentration- and genotype-dependent effects on maize seedling growth and antioxidant responses, highlighting their potential application in sustainable crop production. Further studies are recommended to determine the optimum FeNP concentration for maximizing plant performance while minimizing potential phytotoxic effects.

Introduction

Maize, scientifically known as *Zea mays* L., Zea mays L. is an annual, monoecious cereal crop belonging to the family Poaceae. It has a fibrous root system, an erect unbranched stem, and long leaves with parallel venation. The plant bears separate male (tassel) and female (ear) flowers on the same plant, producing kernels after successful pollination. Morphological traits such as plant height, leaf number, and biomass are important indicators of growth and productivity.

These features have been the subject of numerous studies aimed at understanding the genetic, physiological, and agronomic aspects of maize plants. Maize seed typically includes a hard outer layer called the seed coat, which protects the embryo within (Halli et al., 2021).

Iron (Fe) is an essential micronutrient for plant growth and development, playing a crucial role in various cellular functions such as photosynthesis, respiration, and DNA synthesis (Liu et al., 2022; *Tsai* and Schmidt*, 2020;* Wu and Ling*,* 2019*)*. Despite its relative abundance in the Earth's crust, iron availability for plants is often limited due to its poor solubility in soil, especially in neutral and basic soils (Bakirbas and Walker, 2022; Herlihy et al., 2020). Iron deficiency in plants can lead to leaf chlorosis, characterized by yellowing of leaves due to insufficient chlorophyll production (He et al., 2019; Merry et al., 2022). This deficiency significantly impacts plant vigor and agricultural productivity (Fu et al., 2017; Kong et al., 2022).

Iron deficiency in maize (*Zea mays*) plants can have detrimental effects on their growth and physiological development, ultimately impacting crop yield and quality. Iron is vital for numerous physiological processes, including photosynthesis, respiration, and enzyme activation. Iron deficiency in maize leads to chlorosis, stunted growth, reduced root development, and decreased biomass accumulation (Jalali et al., 2017; Chen et al., 2015). Consequently, maize plants exhibit poor plant vigor and decreased tolerance to environmental stresses, adversely affecting productivity (Soppi, 2018; Garcìa-Casal et al., 2018).

Plants have evolved adaptive mechanisms to enhance iron acquisition from the soil, such as the regulation of iron uptake, transport, and homeostasis (Naranjo-Arcos et al., 2017; Park et al., 2019; Yang et al., 2014). These include the regulation of iron homeostasis through various proteins and transcription factors (Hsiao et al., 2017; Montejano-Ramírez & Valencia-Cantero, 2023; Müller et al., 2015). Additionally, plants can alter their gene expression in response to iron deficiency, as demonstrated by differential physiological responses observed under such conditions (Kallala et al., 2018).

Previous studies on iron nanoparticles have demonstrated their potential as novel tools for improving plant growth and development. Iron oxide nanoparticles have been investigated as potential iron fertilizers to replace traditional Iron fertilizers, addressing various shortcomings associated with conventional fertilizers (Rui et al., 2016). The impact of magnetite nanoparticles coated with aspartic acid on maize seedlings has shown improvements in growth, enzymatic activity, and chlorophyll content (Răcuciu et al., 2022). Furthermore, bare and polyethylene glycol-coated FeO nanoparticles have been observed to enhance seedling growth in *Phaseolus vulgaris L.* (Duran et al., 2018).

To address iron deficiency in maize plants, various strategies were implemented, including soil amendments with iron-containing fertilizers, foliar applications of iron chelates, and breeding for maize varieties with enhanced iron uptake and utilization capabilities (Ramírez et al., 2013). Additionally, improving soil health and pH levels can help enhance iron availability to plants. The study aims to investigate the effects of iron nanoparticles on maize seedlings, evaluating their potential to enhance seed germination, seedling growth, and physiological processes related to plant development.

Understanding the effect of iron nanoparticles on maize seedlings can provide valuable insights into their potential as novel tools for improving agricultural practices and crop productivity. The proposed research will explore the application of iron nanoparticles, which have shown promise in enhancing seed germination, seedling growth, and overall plant productivity, thereby offering a new approach to addressing iron deficiency in maize and other crops.

The goals of this study were to evaluate the effects of iron nanoparticles on the germination of maize seedlings, to evaluate the effect of iron nanoparticles on the growth and biochemical responses of maize seedlings, and to determine the best-performing genotype among the three maize varieties treated with various concentrations of FeNPs.

Materials and Methods

Experimental Site

The experiment was conducted at the Botanical Garden of the Department of Plant Science and Biotechnology, Bayero University, Kano, Nigeria (11.9804° N, 8.4277° E).

Seed Collection

Three maize (*Zea mays L.*) genotypes, namely ZAMMAS27, ZAMMAS51, and a local variety, were obtained from the International Institute of Tropical Agriculture (IITA), Kano Station, Nigeria.

Nano-Particle synthesis

Iron nanoparticles (FeNPs) were synthesized in the Department of Pure and Industrial Chemistry, Bayero University, Kano, following the method described by Huber et al. (2005) with slight modifications. Briefly, 10.138 g of ferric chloride ($FeCl_3$) was dissolved in 250 mL of distilled water to prepare a 0.5 M solution, while 34.50 g of ferrous sulfate heptahydrate ($FeSO_4{\cdot}7H_2O$) was dissolved in 250 mL of distilled water to prepare a 0.25 M solution. Equal volumes (100 mL each) of the ferric and ferrous solutions were mixed in a 500 mL beaker and heated to 60–70°C with continuous stirring. Ammonium hydroxide (NH_4OH) was added dropwise until a dark brown precipitate formed. The suspension was allowed to stand for 24 h to facilitate complete precipitation before filtration to obtain the synthesized FeNPs.

Following synthesis and filtration, the iron nanoparticles were re-dispersed in distilled water to prepare a stock suspension with a concentration of 100 mg/L. The stock suspension was thoroughly homogenized using continuous stirring before preparing working suspensions of 75 mg/L and 50 mg/L by serial dilution with distilled water. All nanoparticle suspensions were freshly prepared prior to seed treatment to ensure uniformity and minimize particle aggregation.

Experimental Treatment

The experiment consists of four Iron Nanoparticles FeNP treatment; 0mg/L (control), 50mg/L, 75mg/L and 100mg/L. Three maize *Zea mays L.* varieties (ZAMMAS27, ZAMMAS51, and Local Variety) were evaluated under each treatment prior to sowing, healthy seeds of each variety were primed separately in the respective FeNP suspensions for 24hrs while the seeds in the control treatment were soaked in distilled water. The experiment was arranged in Randomized complete block design (RCBD) with 3 replicates, each treatment Variety combination was represented by 3 polythene bags resulting in 12 treated combinations (4 FeNP concentrations × 3 maize varieties), 36 experimental ujits (polythene bags) and a total of 108 seeds.

Seed Sowing

Following seed priming, three treated seeds were sown in each polythene bag containing a mixture of topsoil, river sand, and organic manure (2:2:1). Each treatment–variety combination consisted of three replicated polythene bags. The seedlings were watered daily under uniform environmental conditions, and observations were recorded 20 days after sowing.

Data recorded

The evaluated parameters included germination percentage, plant height, number of leaves, chlorophyll content, shoot fresh weight, root fresh weight, shoot dry weight, root dry weight, and superoxide dismutase (SOD) activity.

Plant height was measured from the soil surface to the tip of the tallest leaf using a measuring tape. The number of leaves was determined by direct counting. Chlorophyll content was measured using a SPAD-502 Plus Chlorophyll Meter (Konica Minolta, Inc., Osaka, Japan). Shoot and root fresh weights were determined immediately after harvest using an analytical balance (insert the actual model and manufacturer if known). The plant samples were subsequently oven-dried to a constant weight before determining the shoot and root dry weights. Superoxide dismutase (SOD) activity was determined as described in the next section.

Determination of Superoxide dismutase (SOD) Activity

Superoxide dismutase (SOD) activity was determined at the Faculty of Agriculture, Ahmadu Bello University, Zaria, using a commercially available Superoxide Dismutase Assay Kit (Cayman Chemical Company, Ann Arbor, MI, USA). Plant tissue extracts were prepared according to the manufacturer's instructions. The assay mixture consisted of 50 μL reaction buffer, 50 μL plant extract, and 50 μL enzyme reagent. The reaction mixtures were incubated at room temperature for 30 min, after which absorbance was measured at 450 nm using a spectrophotometer. Superoxide dismutase (SOD) activity was determined using an ELISA Plate Reader (DIA2000, BIOBASE Biodustry (Shandong) Co., Ltd., Jinan, Shandong Province, China). SOD activity was determined according to the manufacturer's instructions. Enzyme activity was calculated from the standard calibration curve provided with the assay kit and expressed as units per milligram of protein (U mg^{-1} protein) (Răcuciu et al., 2022).

Data Analysis

Experimental data were analyzed using one-way analysis of variance (ANOVA). Mean separation was performed using the Least Significant Difference (LSD) test at the 5% probability level ($P < 0.05$). Statistical analyses and graphical presentations were carried out using Minitab version 18, GraphPad Prism version 9, and R software version 4.0.1.

Results

Descriptive Statistics of the Nine Evaluated Traits of Three Varieties of Maize

Table 1 summarizes the descriptive statistics for the nine evaluated growth and biochemical traits of the three maize genotypes. Superoxide dismutase (SOD) activity recorded the highest mean value (54.02 ± 20.49), whereas root dry weight (RDW) had the lowest mean (0.12 ± 0.06). Root fresh weight (RFW) exhibited the highest coefficient of variation (79.94%), indicating considerable variability among treatments, while plant height (PHT) showed the lowest coefficient of variation (24.50%), suggesting relatively stable responses across treatments.

Effects of Different Concentrations of Nanoparticles and Varieties on Growth and Biochemical Parameters in Maize

The main effect of iron Nanoparticles concentrations and maize variations on the evaluated growth and biochemical parameters are presented in Table 2. Application of iron nanoparticles significantly influenced most of the evaluated growth and biochemical parameters. Germination percentage, number of leaves, plant height, shoot fresh weight, root fresh weight, root dry weight, and superoxide dismutase activity differed significantly among FeNP concentrations and maize varieties ($P < 0.05$). Chlorophyll content also varied significantly among treatments, indicating that FeNP application affected photosynthetic pigment accumulation in a concentration-dependent manner. However, shoot dry weight showed comparatively smaller treatment effects.

Effect of Iron Nanoparticles on Plant Height (PHT) of Three Varieties of Maize

As shown in Table 2, Plant height was significantly influenced ($P < 0.05$) by FeNP concentration. The highest mean plant height (13.74 cm) was recorded at 100 mg/L, followed by 75 mg/L (12.51 cm), the control (12.22 cm), while the lowest mean value (10.96 cm) was observed at 50 mg/L. Among the maize varieties, the Local Variety produced the tallest plants (13.83 cm), followed by ZAMMAS51 (13.08 cm), whereas ZAMMAS27 recorded the lowest mean plant height (10.17 cm). These findings indicate that maize genotypes responded differently to FeNP application.

Effect of Iron Nanoparticles on Number of Leaves (NLF) of Three Varieties of Maize

According to Table 2, the number of leaves differed significantly ($P < 0.05$) among the FeNP concentrations. The control treatment produced the highest mean number of leaves (5.33), followed by 75 mg/L (3.11), 50 mg/L (3.33), whereas the lowest mean value (2.22) was recorded at 100 mg/L. Across the maize varieties, ZAMMAS51 produced the highest mean number of leaves (3.75), followed by the Local Variety (2.50), while ZAMMAS27 recorded the lowest value (2.50). ZAMMAS27 and the Local Variety both have 2.50 leaves. Hence, they were not significantly different.

Effect of Iron Nanoparticles on Shoot Fresh Weight (SFW) of Three Varieties of Maize

As presented in Table 2, Shoot fresh weight was significantly affected ($P < 0.05$) by FeNP concentration. The highest mean shoot fresh weight (26.48 g) was recorded at 100 mg/L, followed by 75 mg/L (22.13 g), 50 mg/L (14.90 g), whereas the control treatment produced 17.57 g. Among the maize varieties, ZAMMAS27 recorded the highest mean shoot fresh weight (23.03 g), followed by the Local Variety (20.51 g), while ZAMMAS51 produced the lowest value (17.27 g).

Effect of Iron Nanoparticles on Root Fresh Weight (RFW) of Three Varieties of Maize

Table 2 shows that Root fresh weight varied significantly ($P < 0.05$) among FeNP concentrations. The highest mean root fresh weight (7.13 g) was recorded in the control treatment, followed by 75 mg/L (3.83 g), 50 mg/L (7.12 g), whereas the lowest mean value (3.00 g) was observed at 100 mg/L. Among the maize varieties, ZAMMAS27 produced the highest mean root fresh weight (11.43 g), followed by the Local Variety (2.58 g), while ZAMMAS51 recorded the lowest value (1.81 g).

Effect of Iron Nanoparticles on Shoot Dry Weight (SDW) of Three Varieties of Maize

According to Table 2, significant differences ($P < 0.05$) in shoot dry weight were observed among the FeNP concentrations. The highest mean shoot dry weight (1.226 g) was recorded at 75 mg/L, followed by the control (0.837 g), 100 mg/L (0.666 g), whereas the lowest value (0.571 g) was obtained at 50 mg/L. Across the maize varieties, the Local Variety recorded the highest mean shoot dry weight (1.002 g), followed by ZAMMAS51 (0.779 g), while ZAMMAS27 recorded the lowest value (0.695 g).

Effect of Iron Nanoparticles on Root Dry Weight (RDW) of Three Varieties of Maize

As presented in Table 2, Root dry weight differed significantly ($P < 0.05$) among the FeNP concentrations. The highest mean root dry weight (0.142 g) was obtained at 75 mg/L, followed by 100 mg/L (0.086 g), whereas the lowest value (0.080 g) was recorded at 50 mg/L. Among the maize varieties, ZAMMAS27 produced the highest root dry weight (0.155 g), followed by ZAMMAS51 (0.141 g), while the Local Variety recorded the lowest value (0.049 g).

Effect of Iron Nanoparticles on Chlorophyll Content (CLC)

Chlorophyll content was significantly influenced ($P < 0.05$) by FeNP concentration as shown in Figure 1. The control treatment produced the highest mean chlorophyll content (32.78), whereas the lowest value (23.24) was recorded at

50 mg/L. Chlorophyll content increased to 26.25 at 75 mg/L and further to 29.49 at 100 mg/L, indicating that the response was not strictly concentration-dependent. Among the maize varieties, the Local Variety recorded the highest mean chlorophyll content (29.13), followed by ZAMMAS27 (28.62), while ZAMMAS51 had the lowest value (26.08).

Effect of Iron Nanoparticles on Percentage Germination (%GMN) of Three Varieties of Maize

Percentage germination was significantly influenced by FeNP concentration ($P < 0.05$). The control treatment recorded the highest mean germination percentage (33.33%), followed by 50 mg/L (19.25%), 75 mg/L (14.82%), whereas the lowest germination percentage (9.63%) was recorded at 100 mg/L. Among the maize varieties, ZAMMAS51 recorded the highest mean germination percentage (21.67%), followed by ZAMMAS27 (20.56%), while the Local Variety recorded the lowest value (15.51%), Figure 2.

Effect of Iron Nanoparticles on Superoxide Dismutase (SOD) of Three Varieties of Maize

Figure 3 shows the response of Superoxide dismutase (SOD) activity to iron nanoparticles treatment. The SOD increased significantly ($P < 0.05$) with increasing FeNP concentration. The highest mean SOD activity (65.95 U mL^{-1}) was observed at 100 mg/L, followed by 75 mg/L (54.83 U mL^{-1}), 50 mg/L (51.76 U mL^{-1}), while the control treatment recorded the lowest activity (43.52 U mL^{-1}). Among the maize varieties, ZAMMAS51 exhibited the highest mean SOD activity (74.65 U mL^{-1}), followed by ZAMMAS27 (56.00 U mL^{-1}), whereas the Local Variety recorded the lowest activity (31.41 U mL^{-1}).

Pearson Correlation Analysis of Nine Evaluated Traits of Three Varieties of Maize

The relationships among the evaluated traits are presented in Figure 4. Pearson correlation analysis revealed moderate positive correlations between percentage germination and number of leaves ($r = 0.65$; $P < 0.001$) and between plant height and shoot dry weight ($r = 0.29$). Root dry weight showed positive correlations with root fresh weight ($r = 0.48$; $P < 0.01$) and SOD activity ($r = 0.47$; $P < 0.01$). Significant negative correlations were observed between plant height and root fresh weight ($r = -0.56$; $P < 0.001$) as well as between plant height and root dry weight ($r = -0.45$; $P < 0.001$). Number of leaves also showed a weak negative correlation with root dry weight ($r = -0.42$; $P < 0.001$).

Table 1: Descriptive Statistics of Nine Evaluated Traits of Three Genotypes of maize

Traits	Mean	SD	CV (%)	Range	Skewness	Kurtosis
%GMN	19.26	11.24	58.38	31.15	0.12	-1.39
NLF	3.50	2.17	62.04	8.00	0.11	-0.74
PHT	12.36	3.03	24.50	11.50	-0.34	-0.40
CLC	27.94	7.66	27.41	30.30	-0.60	-0.27
SFW	20.27	8.81	43.47	26.64	0.58	-1.21
RFW	3.61	2.88	79.94	8.70	0.94	-0.74
SDW	0.83	0.46	55.93	1.48	1.03	-0.40
RDW	0.12	0.06	51.42	0.19	-0.10	-1.27
SOD	54.02	20.49	37.93	72.74	0.36	-0.54

Key: SD = Standard Deviation, CV= Coefficient of Variance.

Table 2: Effects of Different Concentration of Iron nano Particles and Varieties on Growth and Biochemical Parameters on Maize.

Factors	Traits								
Conc. (%)	%GMN	NLF	PHT	CLC	SFW	RFW	SDW	RDW	SOD
Control (0)	33.330[a]	5.333[a]	12.222[a]	32.777[a]	17.571[c]	7.132[a]	0.837[b]	0.102[c]	43.522[d]
50	19.253[b]	3.333[b]	10.955[ab]	23.244[c]	14.902[d]	7.116[a]	0.571[d]	0.130[b]	51.763[c]
75	14.818[c]	3.111[b]	12.511[ab]	26.248[bc]	22.133[b]	3.833[b]	1.226[a]	0.142[a]	54.832[b]
100	9.630[d]	2.222[b]	13.744[b]	29.488[ab]	26.475[a]	3.003[c]	0.666[c]	0.086[d]	65.954[a]
Variety									
ZAMMAS51	21.666[a]	3.750[a]	13.075[a]	29.125[a]	17.270[c]	1.809[c]	1.002[a]	0.141[b]	74.645[a]
ZAMMAS27	20.555[b]	2.500[b]	10.166[a]	28.616[ab]	23.033[a]	11.425[a]	0.695[c]	0.155[a]	56.001[b]
LOCAL VARIETY	15.551[c]	4.250[a]	13.833[b]	26.078[c]	20.507[b]	2.580[b]	0.779[b]	0.049[c]	31.406[c]

Key: Percentage Germination (%GMN), Number of Leaves (NLF), Plant Height (PHT), Chlorophyll Content (CLC), Shoot Fresh Weight (SFW), Root Fresh Weight (RFW), Shoot Dry Weight (SDW), Superoxide Dismutase (SOD). Means with the same letters are not statistically significant at P<0.05

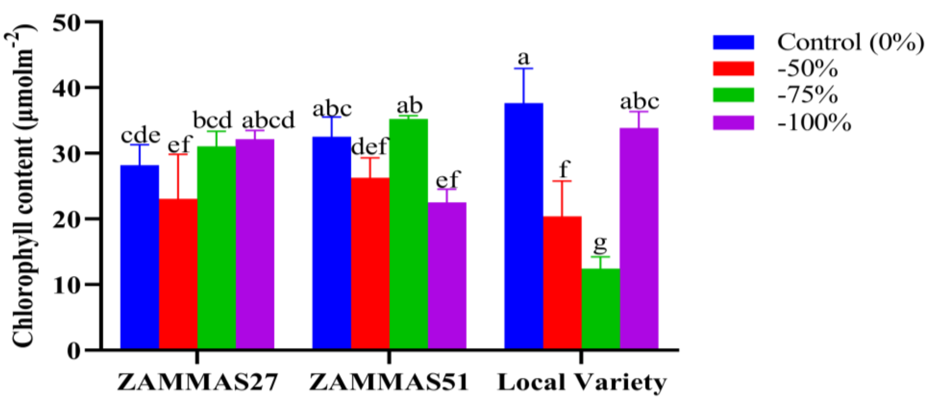

Figure 1: Effects of different concentrations of FeNPs on the chlorophyll content of three genotypes of maize. Data with the same letters are not statistically significant at P<0.05

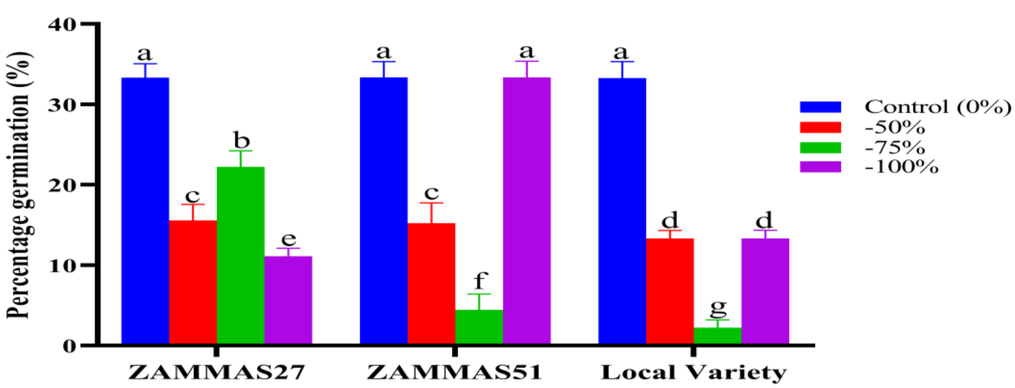

Figure 2: Effects of different concentrations of FeNPs on the percentage germination of three genotypes of maize. Data with the same letters are not statistically significant at P<0.05

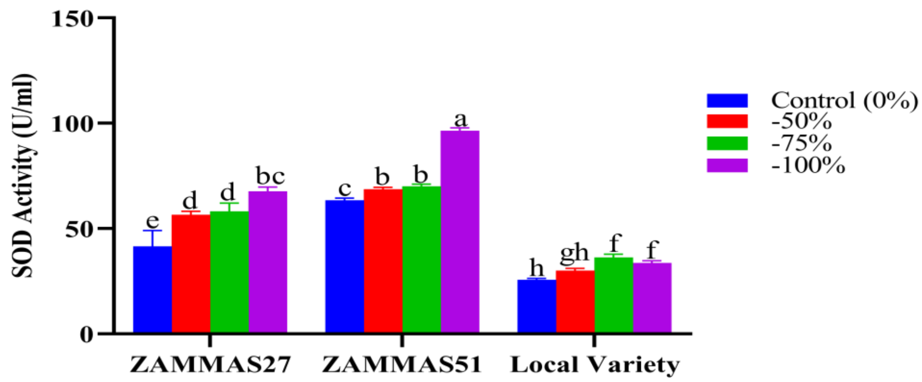

Figure 3: Effects of different concentrations of FeNPs on the SOD activity of three genotypes of maize. Values are the means of three replicates ± standard deviation. Data with the same letters are not statistically significant at P<0.05

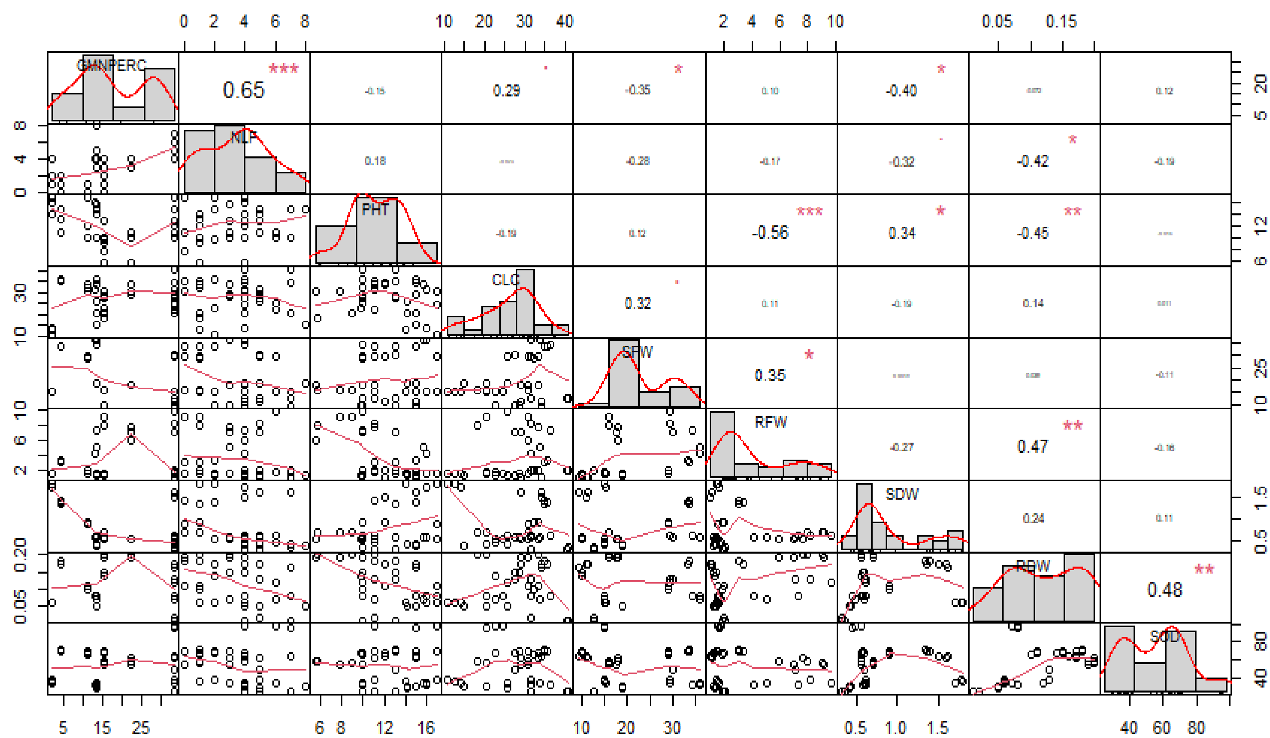

Figure 4: Correlation analysis of nine traits. Along the diagonal shows the traits and the frequency histogram. Below the diagonal indicate the scatter plot and above the diagonal indicate the Pearson correlation coefficient indicated. Key points: * = P<0.05, ** = P<0.01, and *** = P<0.001. PHT = Plant height, NLF = Number of Leaves, SFW = Shoot Fresh Weight, RFW = Root Fresh Weight, SDW = Shoot Dry Weight, RDW = Root Dry Weight, SOD = Superoxide dismutase, CLC = Chlorophyll content, GMNPERC = Percentage Germination

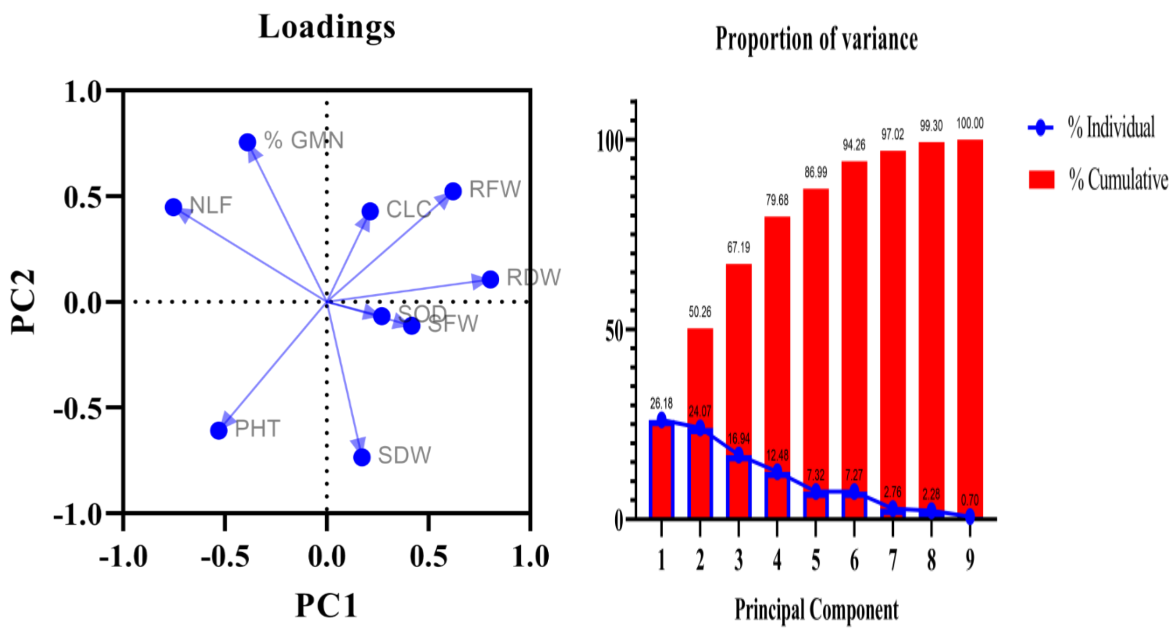

Figure 5: Principal component analysis of nine traits. (A) Displays the loading plots of the two principal components. (B) Depicts the individual and cumulative contribution of each principal component.

Principal Component Analysis (PCA) of Nine Evaluated Traits of Three Maize Varieties

The PCA biplot and proportion of variance explained are presented in Figure 5. Principal component analysis indicated that the first two principal components explained approximately 50.23% of the total variation among the evaluated traits. Principal Component 1 (PC1) accounted for 26.16%, while Principal Component 2 (PC2) explained 24.07% of the total variation. Root fresh weight, chlorophyll content, and root dry weight exhibited positive loadings on the positive side of PC1, whereas plant height, number of leaves, and percentage germination were negatively associated with PC1. Superoxide dismutase activity and shoot fresh weight clustered together, suggesting a positive association between antioxidant activity and vegetative biomass.

Discussion

Iron is an essential micronutrient required for several physiological and biochemical processes in plants, including chlorophyll biosynthesis, photosynthesis, respiration, electron transport, and antioxidant defense. Consequently, an adequate iron supply is critical for normal plant growth and development. The present study demonstrated that iron nanoparticles (FeNPs) significantly influenced the germination, growth, chlorophyll content, and antioxidant activity of maize seedlings, although the responses varied with nanoparticle concentration and genotype. These findings support previous reports that iron nanoparticles can modify plant physiological and biochemical processes depending on the applied concentration and plant species (Liu et al., *2022;* Tsai & Schmidt, *2020;* Wu & Ling, *2019*).

The reduction in germination percentage observed at higher FeNP concentrations suggests that excessive nanoparticle exposure may inhibit seed germination by interfering with water uptake, disrupting metabolic activities, or inducing oxidative stress during the early stages of seedling development. Similar inhibitory effects of high concentrations of iron nanoparticles have been reported in maize and other crop species, where excessive nanoparticle accumulation reduced seed germination and early seedling growth (Rui et al., 2016; Duran et al., 2018).

Growth responses, including plant height, leaf production, and biomass accumulation, differed among the evaluated maize genotypes, indicating genotype-dependent responses to FeNP application. Moderate FeNP application, particularly at 75 mg/L, promoted greater shoot and root dry matter accumulation, suggesting that optimum nanoparticle concentrations enhance nutrient utilization and biomass production more effectively than either lower or higher concentrations. Similar genotype-specific responses to nanoparticle application have been reported in cereal crops and other agricultural species (Răcuciu et al., 2022; Rui et al., 2016).

Superoxide dismutase (SOD) activity increased progressively with FeNP concentration, with the highest activity recorded at 100 mg/L. This increase indicates activation of the antioxidant defense system in response to nanoparticle-induced oxidative stress. Superoxide dismutase serves as the first line of enzymatic defense against reactive oxygen species by catalyzing the conversion of superoxide radicals into hydrogen peroxide and molecular oxygen. Similar increases in antioxidant enzyme activity following nano-iron application have been reported by Jalali et al. (2017), who concluded that iron nanoparticles enhance plant tolerance to oxidative stress by stimulating antioxidant defense mechanisms.

Chlorophyll content responded differently across FeNP concentrations. Although the control treatment produced the highest chlorophyll content, the reduction observed at 50 mg/L was followed by a gradual increase at 75 mg/L and 100 mg/L, indicating a non-linear response rather than a continuous decline. This suggests that the physiological effects of FeNPs on chlorophyll biosynthesis depend on the concentration applied. Similar concentration-dependent responses have been reported by Rui et al. (2016), although variations among plant species and experimental conditions have also been documented (Duran et al., 2018).

The positive association between SOD activity and shoot fresh weight suggests that enhanced antioxidant capacity may contribute to improved vegetative growth under FeNP treatment. Likewise, principal component analysis demonstrated close associations between antioxidant activity and biomass-related traits, indicating that efficient oxidative stress management contributes to improved plant performance under nanoparticle application. These findings agree with previous reports that improved antioxidant capacity enhances stress tolerance and promotes plant growth under nanoparticle treatments (Jalali et al. 2017).

Overall, the findings demonstrate that the effects of iron nanoparticles are concentration-dependent and genotype-dependent. Appropriate FeNP concentrations can improve antioxidant activity and biomass production, whereas excessive application may reduce germination. Therefore, careful optimization of FeNP concentration is necessary before recommending their large-scale use as nano-fertilizers for sustainable maize production.

Conclusion

This study demonstrated that iron nanoparticles (FeNPs) significantly influenced the germination, growth, chlorophyll content, and antioxidant activity of three maize (Zea mays L.) genotypes. The effects of FeNPs varied with concentration and genotype. The control treatment (0 mg/L) produced the highest overall germination percentage and chlorophyll content, whereas increasing FeNP concentrations enhanced superoxide dismutase (SOD) activity, with the highest activity observed at 100 mg/L. Shoot and root dry weights were maximized at 75 mg/L, indicating that moderate FeNP application promoted biomass accumulation more effectively than lower or higher concentrations. The three maize genotypes also differed in their responses, with ZAMMAS51 exhibiting the highest antioxidant response, while the Local Variety generally produced greater shoot dry weight and chlorophyll content.

Overall, the findings indicate that iron nanoparticles can improve certain physiological and biochemical characteristics of maize, particularly antioxidant defense and biomass production, when applied at appropriate concentrations. However, excessive concentrations may reduce germination and do not consistently improve all growth parameters. Therefore, the use of FeNPs as nano-fertilizers should be optimized according to concentration and genotype to maximize their benefits while minimizing undesirable effects. This study provides valuable baseline information for the development of iron nanoparticle-based nutrient management strategies in maize and contributes to the growing application of nanotechnology for sustainable crop production.

Recommendations

Further studies should focus on other biochemical markers such as CAT, POD, and protein content to understand the broader effects of iron nanoparticles on maize seedlings. Additionally, the impact of FeNPs on maize plants at maturity and on other cereals like sorghum and millet should be investigated. Molecular genetic analysis is also recommended to explore the effects of FeNPs on maize and other cereals.

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Conflict of interest

The authors declare that there are no financial, commercial, or personal relationships that could have influenced the work reported in this manuscript. Therefore, the authors declare no conflict of interest.

Author contribution

Mustapha Abdulmalik conceived and designed the study, conducted the experiment, collected and analyzed the data, interpreted the results, and prepared the original manuscript. Sani Ibrahim supervised the research, reviewed the methodology, interpreted the findings, and critically revised the manuscript for important intellectual content. The remaining authors contributed to the study design, data interpretation, manuscript review, and approved the final version for publication. All authors read and approved the final manuscript.

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