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Research Article

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EXOGENOUS APPLICATION OF IRON ANDZINC NANOPARTICLES ON GERMINATION AND GROWTH CHARACTERISTICS OF SUGARCANE (*SACCHARUM OFFICINARUM L.*) BUDNODE

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ABSTRACT

The applications of nano-particles (NPs) in agriculture, such as nano-fertilizers, nano-insecticides, and nano-herbicides, are significantly impacted by their specific structure. In an experiment conducted at the College of Agriculture, University of Sargodha, the presence of Fe and Zn nano-particles at different concentrations was investigated to promote the appearance and growth of sugarcane buds. The experiment was conducted using a Randomize Complete Block Design (RCBD) method, with three replications of plant height at different concentrations of Fe NPs and Zn NPs. The results showed that high Zn concentrations, such as 75 and 100 mg L-1, significantly influenced germination-related characteristics, including minimum plant height. Sugarcane buds treated with Fe NPs at 50 mg L-1 and Zn NPs at 100 mg L-1 had the largest leaf area, while buds treated with Zn NPs at 50 mg L-1 had the minimum leaf-to-plant ratio. The topical application of Fe NPs and Zn NPs to sugarcane increased chlorophyll concentration and photosynthetic rate by 1.3 cm. The plant also showed the highest amount of zinc. At 100 mg L-1, the shoot Fe 6.9 concentration in Zn NPs was the highest. In conclusion, adding Zn and Fe nano-particles in amounts ranging from 100 mg L-1 to 50 mg L-1 significantly improved the growth and development of sugarcane bud nodes.

Key words: Sugarcane, germination, growth, yield, Fe, Zn, nanoparticles.

INTRODUCTION

Sugarcane holds significant importance as a cash crop in Pakistan, particularly sugar related industries. Sugarcane contributes 3.4% to the value addition and 0.7% to the GDP. Sugarcane was cultivated on 1.16 million hectares, resulting in a total production of 81 million tons (Economic Survey of Pakistan 2020-21). However, the average production per hectare stood at 69.53 tons. The total annual production of sugarcane in Pakistan has shown significant improvement over the years, increasing from 23.2 million tons in 1971 to 81 million tons in the 2020-21. However, despite these advancements, Pakistan's average production levels still lag behind those of other countries (Raza *et al*., 2023). The sugarcane in Pakistan which indicates low yield rate in this country as compared to other countries. Some of the factors limiting plant growth and yield include high input costs, traditional practices of planting; setting low quality seed, weed burden, pest and disease pressure, poor fertilizer application rates, and conventional methods of sowing (Raza *et al*., 2021). Out of these, the plant bed method and quality of major constraints to production in the traditional systems. As noted, Khaliq *et al*. (2020) healthy seeds are significant determinants of the good yields in sugarcane production, and they are a vital input constituting 20% of the production cost. How nanotechnology can help revolutionize the agricultural production system towards its vision and goal (Lowry *et al*., 2019). Many nanoparticles (NPs) and nanomaterials have been synthesized and employed as resources in agricultural field (Shakiba *et al*., 2020). In comparison to conventional pesticides and fertilizers, nanoparticle fertilizers and pesticides not only need less application, but they also contribute to higher crop output and quality (Malik *et al*., 2020). Nanoparticles (NPs) have become popular in crop production and protection as they have diverse applications (Altaeik *et al*., 2023). Impact on seeds and plants including promotion of seed germination, seedling growth, general plant development through manipulating signaling pathways (Mehdi *et al*., 2024). Nanoparticles are small and thus can pass through cellular barriers in a highly efficient manner as stated by Hu *et al* (2020) micronutrients are crucial in the growth, development, and production of viable plant organs for growth. Significantly, iron is involved in all of the physiological processes that plants go through. The following enzymes have co-factor Fe, one of the most significant microelements involved in the catalytic activity of many enzymes: cytochrome and Fe (ll)/2 oxoglutarate-dependent oxygenase. Fe stimulates the production of chlorophyll during photosynthesis and respiration (Majeed *et al*., 2022). Plant growth and development require zinc as well as other minerals. Zinc is necessary for the metabolism of proteins in a number of plant physiological systems Abbasi *et al*. (2023). Zinc oxide enhances seedling vigor, boosts photosynthesis, and helps plants. Furthermore, it performs essential roles in the production of cell membranes, proteins, and harmonies (Itroutwar *et al*., 2020). This Micronutrient deficiency affects the world's soil to a degree of about 30%, which causes issues with the sugar manufacturing system (Savassa *et al*., 2018). Consequently, for high-quality and long-term cane production, both zinc and iron are essential (Mellis*et al*., 2024). The use of nanotechnology for seed foliar dispersion is a relatively new problem, despite earlier studies showing promising outcomes. Foliar spraying has been demonstrated to be the most efficient method of improving crop quality and yield, compensating for nutrient deficiencies, and controlling soil pH. Additionally, it reduces environmental pollution and increases the uptake of nutrients while utilizing less fertilizer. in the soil (Salman *et al*., 2023). Research conducted by Zhao and colleagues (2017) discovered that while the cuticles of leaves allow for gas exchange, they also keep contaminants from entering the leaf's interior. Of course! Here is an updated, non-plagiarized version of the statement. Aguilar *et al*. (2023) studied the effects of nanocoated substances on stomatal penetration when their size exclusion limit was more than 10 nm. Additionally, scientists discovered that by delivering nutrients at exact times and locations, nanocarriers increase the efficiency of nutrient absorption and decrease the quantity of surplus active compounds that are released into the plant system. During the growth season, foliar nutrients are regularly sprayed on agricultural plants at various periods, mostly to supplement root fertilizations. The processes of foliar absorption might be difficult since different leaf surface areas and structures may engage differently in the adoption method depending on factors like species, developmental stage, or environmental circumstances at the time of treatment. Veins, trichomes, stomata, and other epidermal features, as well as the cuticle and cuticular irregularities, allow plants' leaves to absorb nutritional solutions sprayed on them (Fernández *et al*., 2021).

MATERIAL AND METHOD

Site and the soil: The study took place at the Wire House, College of Agriculture, University of Sargodha, Sargodha, Pakistan, from March to July 2022.The research was conducted in Punjab, Pakistan. Sargodha district, characterized by subtropical to semi-arid climatic conditions, typically receives an average annual rainfall ranging from approximately 400 to 500 mm. The rainy season, or monsoon, typically occurs from July to September and accounts for 70% of the total precipitation. For the experiment, sugarcane budnodes were grown in earthen pots measuring 25 × 40 cm^{-2}. These pots were filled with sandy clay loam soil, characterized by a bulk density of approximately 1.04 g cm lake and Hartge, 1986). The amount of water that the soil could hold was changed to 33% and 70%.Seven days prior to planting, the soil was weighed, and the pots were filled accordingly (Aitken and McCallum, 1988).

Experimental design and treatments: The experiment was carried out using a Randomize Complete Block design (RCBD)with three replications in March 2022. The study comprised nine treatments, involving various concentrations of Fe NPs (25, 50, 75, and 100 mgL^{-1}), Zn NPs (25, 50, 75, and 100 mg L^{-1}), and a control group. For the exogenous treatments, the Fe NPs and Zn NPs were weighed and added to deionized water to achieve the for each therapy at the two-leaf stage. Each pot contained 5 budnodes. Irrigation was administered according to the crop's specific water requirements. The plants were uprooted after 120 days of sprouting for further analysis.

Table 1. Soil analysis of experimental site

Soil Properties	Values
Clay (g kg^{-1})	290
Silt (g kg^{-1})	255
Sand (g kg^{-1})	455
Texture	Sandy loam
pH	7.4
EC (mS cm^{-1})	0.83
Organic matter (g kg^{-1})	1.32
Available P (mg kg^{-1})	3.17
Saturation (%)	38.20
Available N (mg kg^{-1})	4.12
Available K (mg kg^{-1})	88.10

Preparation of Iron and Zn nanoparticles: In order to make iron nanoparticles, 200 mL of distilled water used to dissolve 20 g of ferrous sulphate heptahydrate (FeSO4•7H2O), which was then vigorously agitated on a magnetic shaker. 10g of ferric chloride hexahydrate (FeCl3•6H2O) were dissolved in one hundred milliliters of distilled water in a different container. After being mixed together in a beaker, these two solutions were set on the magnetic shaker. In the meantime, 125 milliliters of distilled water used to dissolve 15 grammes of sodium hydroxide (NaOH) to 0.3 M solution. Using a pipette, the NaOH solution was gradually added to the iron solution while the magnetic shaker was being used to continuously mix. Up until the point of complete addition, NaOH was added. Filter paper was used to filter the finished solution. The solid product that had accumulated on the filter paper was dried for 48 hours at 70°C in an oven. A mortar and pestle were used to gather the solid residue and dry it into a fine powder. After additional characterization, these iron nanoparticles in powder form were kept for use in other research projects. In this study, the sol-gel method was employed to synthesize zinc oxide (ZnO) nanoparticles. Ethanol was utilized as the solvent to dissolve the source material, zinc acetate dihydrate, or Zn (CH3COO)2•2H2O. More precisely, sodium hydroxide (NaOH) and distilled department was established with the use of water. The synthesized ZnO nanoparticles were characterized using the techniques described by Hasnidawani *et al*. (2016) for nanoparticle analysis, field emission scanning electron microscopy (FESEM), energy-dispersive X-ray spectroscopy (EDX), and X-ray diffraction (XRD).

Statistical analysis: The data collected from the variables for all the parameters were statistically analyzed using Fisher's analysis of variance (ANOVA). The analysis of the comparison of treatment means the Least Significant Difference (LSD) test at a 5% probability level based on the method incorporated by Steel *et al*. (1997).

RESULTS

Sprouting percentage (%): Apparently, the ability to grow new shoots at a short interval and simultaneously increasing the yield and quality as well as profitability among the growers. includes data on the proportion of sugarcane that has sprouted or germinated. It was also observed that Fe NPs and Zn NPs had an insignificant impact on sugarcane. Bud nodes. On the sugarcane bud nodes, nanoparticles elicited an insignificant response. However, this outcome ran counter to what we discovered. The control exhibited the highest number of sprouting sugarcane buds. The findings showed that sugarcane budnodes' capacity to hold onto moisture and offer systemic resistance encouraged sprouting. (Tadu *et al*., 2007). The number of sprouts remained relatively consistent across all treatments. When the p-value was smaller than 0.05, treatments with statistically significant differences were denoted by various letters in a column. The table shows that the control treatment outperformed the other treatments, achieving 100% sprouting. In comparison to the control treatment, the Zn nanoparticle treatment at a dosage of 100 mg L-1 exhibited noticeably greater levels of sprouting. Furthermore, the percentage of sprouting was comparable. Zn NPs were at 25 mg L-1, 50 mg L-1, and 75 mg L-1, whereas Fe NPs were at 25 mg L-1. The therapy involving the use of Fe NPs at a dose of 100 mg L-1 showed the lowest sprouting percentage. Since no chemicals or nanoparticles were used, no statistically significant changes were discovered between the replications of any treatment, notwithstanding the lack of statistical significance in the data.

Table 2. Mean comparison of sprouting % influenced by exogenous application of Fe and Zn NPs on Sugarcane budnode.

Treatments	Means
Control	100
Fe NPs @ 25 mg L^{-1},	73
Fe NPs @ 50 mg L^{-1}	60
Fe NPs @ 75 mg L^{-1}	60
Fe NPs @ 100 mg L^{-1}	53
Zn NPs @ 25 mg L^{-1}	73
Zn NPs @ 50 mg L^{-1}	73
Zn NPs @ 75 mg L^{-1}	73
Zn NPs @ 100 mg L^{-1}	93

Note: Different alphabets in the column confirm significant variation between treatments in accordance with LSD Test ($P \leq 0.05$).

Sprouting index: The indicator of germination or sprouting sugarcane budnodes. The data suggests that the control treatment, Zn NPs, and Fe NPs had basically negligible impacts. While nanoparticles had no discernible influence on sugarcane budnodes, our research showed the opposite. In contrast, a statistically significant maximum sprouting index of 2.3 was obtained with the budnode control treatment. With a Zn NP level of 100 mg L-1, the treatment showed the lowest sprouting index (4.6) Among the treatments whose sprouting index values were statistically similar were Fe nanoparticles at concentrations of 25 mg L-1, 50 mg L-1, 75 mg L-1, and 100 mg L-1, and Zn nanoparticles at concentrations of 25 mg L-1, 50 mg L-1, and 75 mg L-1 (Otto *et al*., 2022). The overall high sprouting index of sugarcane was attributed to the improved budnode sprouting. Crucially, since every treatment was carried out according to the identical protocol—including the control treatment, It sowed without the use of nanoparticles; no discernible differences were seen between the treatments.

Table 3. Mean comparison of sprouting index influenced by exogenous application of Fe and Zn NPs on Sugarcane budnode.

Treatments	Means
Control	2.3
Fe NPs @ 25 mg L^{-1}	3.6
Fe NPs @ 50 mg L^{-1}	3.3
Fe NPs @ 75 mg L^{-1}	4.0
Fe NPs @ 100 mg L^{-1}	3.0
Zn NPs @ 25 mg L^{-1}	3.3
Zn NPs @ 50 mg L^{-1}	3.3
Zn NPs @ 75 mg L^{-1}	2.6
Zn NPs @ 100 mg L^{-1}	4.6

Note: Different alphabets in the column confirm significant variation between treatments in accordance with LSD Test ($P \leq 0.05$).

Time to 50% sprouting (days): The time required for sugarcane buds to reach 50% sprouting. The analysis revealed that 50% of the sugarcane budnodes sprouted after being seeded. Nanoparticles showed non significant response on sugarcane budnodes, The outcome revealed a contradictory reaction. The control treatment showed the longest time to achieve 50% sprouting, whereas sugarcane budnodes treated with Fe nanoparticles at concentrations of 50 mg L^{-1}, 75 mg L^{-1}, and 100 mg L^{-1}exhibited shorter times to reach this milestone compared to the control (Macan *et al*., 2020). On average, Fe nanoparticles at 25 mg L^{-1} took the same time to reach 50% sprouting. that the overall differences were not statistically significant, as the T 50% values did not change much across treatments. Given that the budnodes were naturally planted in controlled environments, no discernible differences were observed. However, the treatment involving Zn nanoparticles at 100 mg L^{-1}demonstrated the shortest time to achieve T50%.

Table 4. Mean comparison of 50% sprouting influenced by exogenous application of Fe and Zn NPs on Sugarcane budnode.

Treatments	Means
Control	11.3
Fe NPs @ 25 mg L^{-1}	14.6
Fe NPs @ 50 mg L^{-1}	12.6
Fe NPs @ 75 mg L^{-1}	12.0
Fe NPs @ 100 mg L^{-1}	12.6
Zn NPs @ 25 mg L^{-1}	13.3
Zn NPs @ 50 mg L^{-1}	13.0
Zn NPs @ 75 mg L^{-1}	13.0
Zn NPs @ 100 mg L^{-1}	13.6

Note: Different alphabets in the column confirm significant variation between treatments in accordance with LSD Test ($P \leq 0.05$).

Mean sprouting time (days): According to the mean sprouting time of sugarcane budnodes was analyzed. Since no nanoparticles were used throughout the investigation, the data showed that there was no discernible difference between the control treatment, Fe nanoparticle therapy, and Zn nanoparticle treatment. Nanoparticles showed nonsignificant response on sugarcane budnodes. However, the control treatment had the maximum mean sprouting time (7.0), indicating that it took the longest time for sprouting to occur (Rehman *et al*., 2021). Treatment, which involved Fe NPs at 50 mg L^{-1}, had a shorter mean sprouting time (4.3) after sowing the budnodes in the soil. Treatments six and seven, named Zn NPs @ 25 mg L^{-1} and Zn NPs @ 50 mg L^{-1} respectively, exhibited a low mean sprouting time compared to other treatments, indicating faster sprouting in these two treatments. Treatment, Zn NPs at 75 mg L^{-1}, had a shorter mean sprouting time (1.6). The treatment that had the highest rate of sprouting was, which involved Fe NPs @ 25 mg L^{-1}, with a minimum sprouting time of Control 0.6. It is important to note that although the observations regarding mean sprouting were non-significant based on the data, indicating no significant difference between the treatments .

Table 5. Mean comparison of mean sprouting time influenced by exogenous application of Fe and Zn NPs on Sugarcane budnode.

Treatments	Means
Control	7.0
Fe NPs @ 25 mg L^{-1}	0.6
Fe NPs @ 50 mg L^{-1}	3.3
Fe NPs @ 75 mg L^{-1}	4.3
Fe NPs @ 100 mg L^{-1}	3.3
Zn NPs @ 25 mg L^{-1}	1.0
Zn NPs @ 50 mg L^{-1}	1.0
Zn NPs @ 75 mg L^{-1}	1.6
Zn NPs @ 100 mg L^{-1}	3.6

Note: Different alphabets in the column confirm significant variation between treatments in accordance with LSD Test ($P \leq 0.05$).

Plant height (cm): Plant height refers to the measurement from the base to the top of each plant. The results indicate that budnodes treated with external application of nanoparticles during the study showed greater plant height compared to untreated budnodes. Although plants treated with Zn nanoparticles at 75 mg L^{-1}were taller than those treated with Fe nanoparticles at 50 mg L^{-1}, their heights were statistically similar. Similarly, the plant height of budnodes treated externally with Fe nanoparticles at 25 mg $L^{-1}$was comparable to plants treated externally with Zn nanoparticles at 50 mg $L^{-1}$and 75 mg L^{-1}. Budnodes treated externally with Fe nanoparticles at 100 mg $L^{-1}$and Zn nanoparticles at 25 mg L^{-1}resulted in plants with statistically similar heights. The minimum plant height observed (37 cm) in the control treatment was statistically equivalent to the height of plants treated externally with Zn nanoparticles at 100 mg L^{-1}. The observed increase in shoot development and the active roles of Zn and Fe in physiological processes such as photosynthesis in plants may explain why foliar application of Zn and Fe nanoparticles at 75 mg $L^{-1}$and 50 mg L^{-1}resulted in increased plant height. Additionally, improved plant growth was observed by Shakuntala *et al*. (2020). Rapid root development is responsible for these beneficial impacts because it increases the capacity for nutrient absorption, transportation, and utilization. This leads to greater growth rates and better plant-water interactions Yu *et al*., 2020).

Table 6. Mean comparison of plant height influenced by exogenous application of Fe and Zn NPs on Sugarcane budnode.

Treatment	**Means**
Control	37 d
Fe NPs @ 25 mg L^{-1}	73 ab
Fe NPs @ 50 mg L^{-1}	83 a
Fe NPs @ 75 mg L^{-1}	63 ab
Fe NPs @ 100 mg L^{-1}	51 c
Zn NPs @ 25 mg L^{-1}	58 abc
Zn NPs @ 50 mg L^{-1}	65 ab
Zn NPs @ 75 mg L^{-1}	81 a
Zn NPs @ 100 mg L^{-1}	42 cd

Note: Different alphabets in the column confirm significant variation between treatments in accordance with LSD test ($P \leq 0.05$).

Leaf area (cm^2): Throughout the investigation, the exogenous administration of Fe and Zn nanoparticles had a substantial effect on the leaf area of sugarcane plants. The plants that were treated with 50 mg $L^{-1}$of exogenous Fe NPs and 100 mg $L^{-1}$of exogenous Zn NPs showed the largest leaf area. There were no differences in the measured leaf area of the plants treated with Fe NPs at concentrations of 25 mg $L^{-1}$and 75 mg L^{-1}, or Zn NPs at concentrations of 25 mg $L^{-1}$and 75 mg L^{-1}. The plants treated with 50 mg $L^{-1}$of exogenous zinc nanoparticles and the untreated plants were found to have the least amount of leaf area. Furthermore, when the height of the plants got increasing dosages of nanoparticles, the research demonstrated that rising concentrations of Fe NPs and Zn NPs proved deleterious to plant growth. The growth of new shoots, which accelerate physiological processes like protein synthesis, photosynthesis, and chlorophyll synthesis, may be the cause of the increased leaf area in plants treated with exogenous Fe NPs at a dosage of 50 mg $L^{-1}$and Zn NPs at a concentration of 75 mg L^{-1}, as reported by Mangrio *et al*. (2020).

Table 7. Mean comparison of leaf area index produced due to endogenous treatment of Fe and Zn NPs exogenously on Sugarcane budnode.

Treatments	Means
Control	1465.9 d
Fe NPs @ 25 mg L^{-1}	3067.3 abc
Fe NPs @ 50 mg L^{-1}	4147.3 ab
Fe NPs @ 75 mg L^{-1}	3058.9 abc

Fe NPs @ 100 mg L^{-1}	2754.2 bcd
Zn NPs @ 25 mg L^{-1}	3552.0 abc
Zn NPs @ 50 mg L^{-1}	2426.5 cd
Zn NPs @ 75 mg L^{-1}	3387.7 abc
Zn NPs @ 100 mg L^{-1}	4295.2 a

Note: Different alphabets in the column verify significant change between treatments in accordance with LSD Test ($P \leq 0.05$).

Number of leaves per plant: External nanomaterial application on the quantity of leaves on a sugarcane plant. In contrast to the results of this investigation, nanoparticles on sugarcane bud nodes caused little reactions. This study demonstrated that the exogenous application had a more significant impact on sugarcane's leaf count; plants that were sprayed through the foliar had the number of most leaves per plant. Thereafter, those plants which were treated by Fe nanoparticles 75 mg $L^{-1}$and Zn nanoparticles 50 mg L^{-1}were found to have more leaves after that. The number of leaves per plant was also similar for the sugarcane budnodes treated exogenously with Fe NPs at 25 mg L^{-1}, 50 mg L^{-1}, and 100 mg L^{-1}, Zn NPs at 25 mg L^{-1}, and 100 mg $L^{-1}$and the control treatment and was not significantly affected. With the increase of Zn and Fe nanoparticles concentration to 100 mg $L^{-1}$the effective number of leaves of the sugarcane plant was reduced. On the other hand, when the study used 75 mg $L^{-1}$of Fe NPs, it discovered that sugarcane budnode sprouting enhanced, sugarcane development was stimulated, shoot length was enhanced, and the leaf area was expanded. These effects resulted in a composition of more leaves. This concurs with the past studies that were conducted by Zhang *et al*. (2015) and Faizan *et al*. (2020) they showed that ZnO and Fe nanoparticles apply in agriculture possess both positive and negative impacts on plant growt

Table 8. Mean comparison of number of leaves per plant influenced by exogenous application of Fe and Zn NPs on Sugarcane budnode.

Treatments	Means
Control	4.6
Fe NPs @ 25 mg L^{-1}	5.0
Fe NPs @ 50 mg L^{-1}	5.3
Fe NPs @ 75 mg L^{-1}	7.0
Fe NPs @ 100 mg L^{-1}	5.6
Zn NPs @ 25 mg L^{-1}	5.3
Zn NPs @ 50 mg L^{-1}	6.0
Zn NPs @ 75 mg L^{-1}	4.3
Zn NPs @ 100 mg L^{-1}	4.6

Note: Different alphabets in the column confirm significant variation between treatments in accordance with LSD Test ($P \leq 0.05$).

Number of tillers: The results regarding the amount of tillers/plant in sugarcane and the effectiveness of foliar nanoparticle spray. As a consequence, the analysis's findings demonstrated that neither Fe NPs nor Zn NPs significantly affected the bud nodes of sugarcane. In light of this, the outcome of the study showed that the application of nanoparticles outside the plants altered the tiller potential of the sugarcane plant; those that received nanoparticles through foliar application had the most numerous tillers per plant on average. The tiller count was then affected by the exogenous administration of two nanoparticles, namely Fe NPs at a dosage of 50 mg L-1 and Zn NPs at a dose range of 25 to 100 mg L-1. Notably, when sugarcane budnodes were exposed to exogenous Fe NPs at concentrations of 25 mg L-1, 75 mg L-1, and 100 mg L-1and the control, there was no discernible change in the number of tillers per plant. Thus, the interaction between water availability and that of nano iron can influence multiplicity of plant growth and development.

Table 9. Mean comparison of a number of tillers per plant influenced by exogenous application of Fe and Zn NPs on Sugarcane budnode.

Treatments	Means
Control	0.3
Fe NPs @ 25 mg L^{-1}	0.3
Fe NPs @ 50 mg L^{-1}	0.6
Fe NPs @ 75 mg L^{-1}	0.3
Fe NPs @ 100 mg L^{-1}	0.3
Zn NPs @ 25 mg L^{-1}	0.6
Zn NPs @ 50 mg L^{-1}	0.6
Zn NPs @ 75 mg L^{-1}	0.6
Zn NPs @ 100 mg L^{-1}	0.6

Note: Different alphabets in the column confirm significant variation between treatments in accordance with LSD Test ($P \leq 0.05$).

Photosynthesis rate (µmol m^{-2} sec^{-1}): The photosynthesis rate of sugarcane increased when sugarcane budnodes were treated exogenously with Fe and Zn nanoparticles. Higher photosynthesis rates were observed in sugarcane buds exposed to Fe nanoparticles (Fe NPs) at 50 mg $L^{-1}$and Zn nanoparticles (Zn NPs) at 75 mg L^{-1}. Sugarcane budnodes treated exogenously with Fe NPs at 25 mg L^{-1}, 75 mg L^{-1}, and 100 mg L^{-1}showed a moderate improvement in photosynthesis rates compared to those treated with Fe NPs at 50 mg L^{-1}, although still higher than the control treatment. Similarly, budnodes treated exogenously with Zn NPs at 25 mg L^{-1}, 50 mg L^{-1}, and 100 mg L^{-1}exhibited lower improvements in photosynthesis compared to those treated with Zn NPs at 75 mg L^{-1}. However, sugarcane plants exhibited the lowest photosynthetic rate (1.310 mol m^{-2} s^{-1}) when they were sowed without exogenous application of budnodes. The increased root penetration and early and vigorous growth that enhance nutrient absorption likely contributed to the higher photosynthesis rate observed in sugarcane plants that were sowed (Zhang *et al*., 2022). Fe and Zinc enhanced the rate of photosynthesis in sugarcane plants because they are essential components of various enzymes and play direct or indirect roles in the formation of proteins, carbohydrates, chloroplasts, and chlorophyll.

Table 10. Mean comparison of photosynthesis rate influenced by exogenous application of Fe and Zn NPs on Sugarcane budnode.

Treatments	Means
Control	1.3 c
Fe NPs @ 25 mg L^{-1}	1.5 b
Fe NPs @ 50 mg L^{-1}	1.6 a
Fe NPs @ 75 mg L^{-1}	1.5 b
Fe NPs @ 100 mg L^{-1}	1.4 ab
Zn NPs @ 25 mg L^{-1}	1.4 ab
Zn NPs @ 50 mg L^{-1}	1.5 b
Zn NPs @ 75 mg L^{-1}	1.6 a
Zn NPs @ 100 mg L^{-1}	1.3 c

Note: Different alphabets in the column confirm significant variation between treatments in accordance with LSD Test ($P \leq 0.05$).

Carotenoids content (µg g^{-1} FW): The data on carotenoids in sugarcane indicated that the application of Fe nanoparticles (Fe NPs) and Zn nanoparticles (Zn NPs) to budnodes resulted in increased levels of carotenoids. Specifically, foliar application of Fe NPs and Zn NPs elevated the carotenoid content in sugarcane leaves, with Fe NPs at 50 mg L^{-1} and Zn NPs at 75 mg L^{-1} showing the highest carotenoid content (78.38 µg g^-1 FW). However, higher concentrations of Fe and Zn nanoparticles led to reduced carotenoid levels compared to the untreated treatment. The control treatment exhibited the lowest carotenoid content (49.62 µg g^-1 FW). This finding aligns with previous studies by Costa*et al*. (2020) both of which reported that nanoparticle treatments resulted in the highest carotenoid concentrations in Oryza sativa and Triticum aestivum plants (Suchowilska *et al*., 2020). The essential roles of Fe and Zn in plant functioning are closely related to the synthesis of carotenoids. As plants experience an increase in chloroplasts, chlorophyll content and photosynthesis rates, they enhance the synthesis of carotenoids (Rai *et al*., 2021).

Table 11. Mean comparison of carotenoids content (µg g-1 FW) influenced by exogenous application of Fe and Zn NPs on Sugarcane budnode.

Treatments	Means
Control	49.62 c
Fe NPs @ 25 mg L^{-1}	71.83 b
Fe NPs @ 50 mg L^{-1}	78.38 a
Fe NPs @ 75 mg L^{-1}	63.66 bc
Fe NPs @ 100 mg L^{-1}	56.70 c
Zn NPs @ 25 mg L^{-1}	58.91 c
Zn NPs @ 50 mg L^{-1}	68.47 abc
Zn NPs @ 75 mg L^{-1}	75.37 ab
Zn NPs @ 100 mg L^{-1}	53.55 c

Note: Different alphabets in the column confirm significant variation between treatments in accordance with LSD Test ($P \leq 0.05$).

Shoot Zn content (mg): Provides insights into the zinc (Zn) content of sugarcane shoots and highlights how exogenous application of Zn and Fe nanoparticles significantly impacted these levels. Nanoparticles showed nonsignificant response on sugarcane budnodes, our result showed contradictory response. However, the data clearly shows significant changes in the concentration of zinc (Zn) in sugarcane shoots because of the exogenous application of zinc (Zn) and iron (Fe) nanoparticles. Maximum Zn level

(25. 30 mg) in sugarcane shoots area was achieved when Zn NPs were applied exogenously at 75 mg L-1. Peculiarly, a high dosage of exogenous Zn NPs led to a general decrease in shoot Zn concentration (17. 10mg) relatively to the control sample (23. 40mg). This might have been because of some interaction of the nutrients, the type of soil, or the effect of some internal changes in the plants. Also, concerning the effect on the shooting Zn content, the presence of an antagonistic interaction between Fe and Zn was observed. The above findings depict that the Fe NPs exogenously applied on sugarcane budnodes caused a higher reduction of the increased shoot Zn content as compared to the control treatment. This interaction between Fe and Zn in the plant nutrition is an antagonistic relationship; a well-documented occurrence (Saenchai *et al*., 2016). It is important to understand how such microelements/phosphates which Zn and Fe interact and depend on each other in the plants to have better control over the nutrients and ability to increase plant growth. When such results are made and conclusions on nutrient management are deduced, then such factors as nutrient availability, plant genetics and climatic conditions must be taken into consideration.

Table 12. Mean comparison of root shoot ratio influenced by exogenous application of Fe and Zn NPs on Sugarcane budnode.

Treatments	Means
Fe NPs @ 25 mg L^{-1}	20.20b
Fe NPs @ 50 mg L^{-1}	21.8 ab
Fe NPs @ 75 mg L^{-1}	20.8 b
Fe NPs at @ 100 mg L^{-1}	19.7 c
Zn NPs @ 25 mg L^{-1}	20.3 b
Zn NPs @ 50 mg L^{-1}	21.4 ab
Zn NPs @ 75 mg L^{-1}	25.30 a
Zn NPs @ 100 mg L^{-1}	17.10 d

Note: Different alphabets in the column confirm significant variation between treatments in accordance with LSD Test ($P \leq 0.05$).

Shoot Fe content (mg): These are the Zn and Fe-NP concentrations on the Fe content of sugarcane shoots that the data represents and shows the impact of various treatment levels on the Fe levels of the shoots during the experiment. It was found in the study that the presence of Fe and Zn nanoparticles had a substantial effect on the shoot's Fe content. Sugarcane budnodes treated with 75 mg L^{-1}Fe NPs had the highest amount of Fe in shoots, but budnodes treated with 50 mg L^{-1}Zn NPs had only slightly lower levels. Notably, the amounts of shot Fe content in these two groups were almost identical. Similarly, plants treated with Zn NPs exogenously at a dose of 25 mg L^{-1}had a considerably similar shoot Fe content to those treated with the control treatment. Even more unexpectedly, the plants' shoot Fe content dropped as iron nanoparticle concentration (Fe NPs at 1000 mg L^{-1}) rose. And this suggests that iron toxicity, a phrase used to characterize circumstances in which the metal is abundant in plants, has an adverse effect on nutritional status and uptake. However, it is important to recognize that these results show how delicate the line is when adjusting nutrients in plants. Similarly, iron is necessary for the growth and development of plants, but excessive quantities can be hazardous. Plant nutrition management requires an understanding of nutrient interactions, absorption methods, and optimal nutrient concentrations (Borges *et al*., 2020).

Table 13. Mean comparison of Zn content influenced by exogenous application of Fe and Zn NPs on Sugarcane budnode.

Treatments	Means
Control	23.40 ab
Fe NPs @ 25 mg L^{-1}	20.20 abc
Fe NPs @ 50 mg L^{-1}	21.80 ab
Fe NPs @ 75 mg L^{-1}	20.80 abc
Fe NPs @ 100 mg L^{-1}	19.70 bc
Zn NPs @ 25 mg L^{-1}	20.30 abc
Zn NPs @ 50 mg L^{-1}	21.40 ab
Zn NPs @ 75 mg L^{-1}	25.30 a
Zn NPs @ 100 mg L^{-1}	17.10 c

Note: Different alphabets in the column confirm significant variation between treatments in accordance with LSD Test ($P \leq 0.05$).

DISCUSSION

A major cash crop in Pakistan and across the world is sugarcane. Traditionally, sugarcane is planted using sets, which require a large quantity. A newer innovative technique involves growing sugarcane through budnodes, when the sugarcane budnodes are cut off from the stalk and planted straight into the field. Nevertheless, budnodes provide difficulties for germination because, in contrast to setts, they dry up more quickly. With the development of different nanoparticles and nanomaterials to improve crop productivity, nanotechnology presents the possibility of a technologically driven revolution in agriculture.

Sugarcane budnodes sprouted and in response to iron nanoparticles (Fe NPs) and zinc nanoparticles (Zn NPs). In the experiment, untreated budnodes were applied with varying doses of Zn NPs (25, 50, 75, and 100 mg L^{-1}) and Fe NPs (25, 50, 75, and 100 mg L^{-1}). Time to 50% sprouting, mean sprouting time, final sprouting percentage, sprouting index, plant height, number of leaves per plant, leaf area per plant, and Zn and Fe content in plants. WhenFe NP concentrations of 100 mg $L^{-1}$and Zn NP concentrations of 50 mg L^{-1}, sugarcane budnodes showed the Maximum shoots. The ultimate plant height showed similar tendencies, with sugarcane reaching its maximum height Fe NPs at 100 mg $L^{-1}$and then Zn NPs at 50 mg L^{-1}. Sugarcane budnodes treated exogenously with zinc NPs at a dose of 100 mg L-1 showed the largest leaf area at 4295.2, whereas budnodes treated with zinc NPs at a dose of 50 mg L-1 showed the least leaf area at 1465.9. The highest number of leaves per plant was seen in the sugarcane budnodes treated with Fe and Zn NPs when the NPs were sprayed foliarly at 75 mg L-1 (4.3) and 50 mg L-1 (6.0), respectively. The application of exogenous Zn NPs produced more tillers, with the greatest root shoot ratio reported at 50 mg L-1 of Zn NPs (8.2), followed by the control treatment (6.8). The findings showed that the foliar application of Fe NPs and Zn NPs enhanced the photosynthetic rate, carotenoid and chlorophyll concentrations of sugarcane. On sugarcane budnodes, a maximum zinc concentration of 75 mg L-1Zn NPs was observed in the plant sections. Likewise, the Fe content in shoots of sugarcane budnodes planted in growth media containing 50 mg $L^{-1}$and 75 mg L^{-1}Zn NPs was also the highest. Therefore, the external use of Zn and Fe NPs at a concentration of 100 mg L^{-1}, and 50 mg L^{-1}positively affected the germination, growth, and development of sugarcane budnodes. Zinc, with the chemical symbol Zn, is another micronutrient, while iron, with the symbol Fe, is a crucial component for better plant growth. By using the enormous surface area to volume ratio of nanoparticles, nanotechnology has produced techniques for the most effective way to supply these nutrients to plants. The influence of exogenous Fe and Zn nanoparticles on the germination and development of sugarcane (Saccharum officinarum L.) budnodes is reviewed in this research, with an emphasis on the beneficial effects and potential cellular mechanisms (Iwuozor*et al*., 2022). This claimed that plant tissues can access Fe and Zn nanoparticles more readily than they can large forms of the metals. Tamez *et al*. (2019) reported that the increased surface area facilitates the easy dissolution and absorption of nutrients by plant cells, hence promoting improved germination. Additionally, it helps lessen abiotic stressors like salt and dryness that could prevent seeds from germinating. Through the ingestion of nanoparticles, the plant's bud nodes receive better nutrition and water, which strengthens the plant's resistance to environmental challenges (Tamashiro *et al*., 2022). Plants can change their levels of reactive oxygen species (ROS) through the action of nanoparticles. Fe and Zn nanoparticles strengthen the plant's antioxidant defence system, reducing oxidative stress and improving plant health in the process (Mellis *et al*., 2024). It has been demonstrated that externally applied Fe and Zn nanoparticles have a tremendous potential to improve sugarcane bud node germination, growth, and development. Their effectiveness can be attributed to their high biologic encompassing power, inductive enzyme activity, and stress protection. However, to ensure the effective deployment of nanoparticle technologies in agriculture, a more thorough examination of the application techniques, toxicity characteristics, and environmental impacts has to be conducted. Future research should focus on the following areas: improving nanoparticle compositions, identifying some long-term effects, and exploring viable sustainable approaches for producing nano-agriculture (Verma *et al*., 2023).

CONCLUSION

Based on our research findings, the application of nanoparticles (Fe NPs at 100 mg L1 and Zn NPs at 50 mg L2) significantly improved seedling growth and germination in sugarcane bud nodes. These nanoparticles have shown great potential for have observed that Fe NPs specifically promote sugarcane growth by increasing plant length, cane weight, and the number of leaves and tillers. enhancing bud node technology, which could lead to better sugarcane seed management for farmers and improve sugarcane cultivation overall. Specifically, Fe NPs have been observed to promote sugarcane growth by increasing plant length, cane weight, and the number of leaves and tillers. Overall, the use of nanoparticles on sugarcane bud nodes has the potential to boost sugarcane productivity

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