IMPACT OF NANOSCALE ZINC OXIDE PARTICLE ON THE GROWTH, YIELD AND SOIL PROPERTIES UNDER AGENCY AREA OF ANDHRAPRADESH

ABSTRACT

Field experiment was carried out on silt clay loam soils at Regional Agricultural Research station, Chintapalle, Visakhapatnam, Andhra Pradesh during 2018-2020 to study the yield of response of rainfed groundnut with different levels of Nano particulate zinc oxide application along with NPK Fertilizers. The experiment consisting of eight treatments, three replications with RDF design. Results of nano particulate zinc oxide on pod yield showed that (24.36 Q/ha) in the RDF+ nano scale zinc oxide level @ 200 ppm at 25 and 45 DAS was increased over to that of normal recommended dose of N,P,K fertilizers (100% RDF) which recorded pod yield of 17.14 Q/ha only. Application of $ZnSO_4$ through soil along with RDF showed good results (17.24 Q/ha) than RDF + Foliar application of $ZnSO_4$ @ 2g/lit at 25 and 45 DAS (20.66 Q/ha). Application of micronutrient (ZnO) had helped in further increase in grain yields at both levels of ZnO (150% and 200% ZnO at 25 and 45 DAS). Among different treatments, significantly higher yield (24.36 Q/ha) was recorded with application of RDF + Foliar application of ZnO @ 200 ppm at 25 and 45 DAS than the only with RDF (17.4 Q/ha). With respect to method of application of ZnSO4 through soil and foliar application ZnSo4 @ 2g/lit at 25 and 45 DAS was found to be higher both levels of RDF (Pod yield of 14.6 Q/ha at RDF + Soil application of ZnSO4 @ 50 kg ha-1 and 17.24 Q/ha at RDF+ foliar application of ZnSO4@ 2g/lit at 25 and 45 DAS). With respect to other plant characteristics, comparatively more plant height (43.53 cm) at RDF + foliar application of ZnO @ 200 ppm at 25 and 45 DAS. Regarding yield attributes significantly higher test weight (30.9 g) were recorded at RDF + 200 ppm ZnO at 25 and 45 DAS. Post –harvest soil sample analysis showed highest availability of nutrients in respect of soil, the results revealed that there was no significance difference among the treatments regarding Avail. N, Available K and pH. The lowest Phosphorus (17.20 kg ha^{-1})

was recorded with RDF+ Foliar application of nanoscale ZnO 200ppm at 25 & 45 DAS and highest (24.46 kg ha^{-1}) was recorded in T_9 (RDF+ Foliar application of nanoscale ZnO 100 ppm at 25 & 45 DAS) .

An investigation was initiated at Department of Soil science, Regional Agricultural Research Station, Chintapalle to examine the effects of nano zinc oxide on Groundnut (*Arachis hypogea* L.) growth, yield and Zn content in Leaves, stem and roots. A field experiment consisted of nine treatments comprised of T_1: control, T_2: RDF, T_3: RDF+ Soil application $ZnSO_4$@ 50 Kg/ha, T_4: RDF+ Foliar application $ZnSO_4$ 2 g/L at 25 & 45DAS, T_5: RDF+ Foliar application of nanoscale ZnO 150 ppm at 25 & 45 DAS, T_6: RDF+ Foliar application of nanoscale ZnO 200 ppm at 25 & 45 DAS, T_7: RDF+ Foliar application of nanoscale ZnO 400 ppm at 25 & 45 DAS, T_8: RDF+ Foliar application of nanoscale ZnO 50 ppm at 25 & 45 DAS, T_9: RDF+ Foliar application of nanoscale ZnO 100 ppm at 25 & 45 DAS

Keywords: Nano particles, zinc oxide, Groundnut, HAT zone , foliar application

Introduction

Zinc (Zn) is typically the second most abundant transition metal in organisms after iron and the only metal represented in all six enzyme classes (oxidoredutases, transferases, hydrolases, lyses, isomerase and ligases (Auld, 2001). Zinc is an essential micronutrient for humans, animals and plants. Higher plants generally absorb Zn as a divalent cation (Zn^{+2}), which acts either as the metal component of enzymes or as a functional structural or a regulatory co-factor of a large number of enzymes. A number of researchers have reported the essentiality and role of zinc for plant growth and yield (Camp and Fudge, 1945; Anderson, 1972: Mengel and Kirkby, 1978; Fageria et al., 2002). Based on analysis of 298 soil samples collected from different countries in the world, Zn deficiency has been found to be the most widespread micronutrient deficiency (Sillanppa, 1990: Sillanpaa and Vlek, 1985). In India, Zn is now considered the fourth most important yield-limiting nutrient after nitrogen (N), Phosphorus (P) and Potassium (P). In India alone, 50% of the soils that groundnut is grown in show Zn deficiency, which is causing considerable yield loss (Singh, 1999). Half of the cultivated soils in

Turkey have Zn deficiency (Eyupoglu et al., 1993) considerable increases in grain yield by Zn application was also demonstared in India (Tandon, 1995). Zinc is required for chlorophyll production, pollen function, fertilization and germination (Kaya and Higgs, 2002: Pandey et al., 2006: Cakmak, 2008). Zinc plays an important role in biomass production (Kaya and higgs 2002). Among the micronutrients, Zn and Mn can affect the susceptibility of plants to drought stress (Khan et al., 2003). A number of mechanisms may underlie Zn deficiency (Rengel, 2001). Depending on experimental conditions and the plant species, the most important mechanisms may be Zn utilization in tissues, called internal efficiency and Zn uptake called external efficiency (Genc et al., 2006). Zinc is intermediate in its mobility or phloem export. Longnecker and Robson (1993) suggested that zinc efficiency depends on the amount supplied and the nature of plant species. Zinc moves from leaves to roots, stem and developing grain and from one root to another (Rengel, 2001). Higher uptakes of other nutreints are also known to increase the demand of Zn.

Graham et al. (2001) reported that over 3 billion people across the world from micronutrient deficiencies and suggested that a considerable amount of research in the 21 st century should be devoted to develop technologies for enhanced uptake and accumulation of micronutrients in edible plant parts. Groundnut is an important legume food crop of India grown in about 8 million ha of land. Groundnut cultivation occurs in 108 countries around the world. The average productivity of groundnut in India around 1178 kg ha^{-1}, which is far less than the world's average 1400 kg ha^{-1}. The low productivity is mainly due to the fact that the crop is mostly grown in rainfed low fertility soils. Micronutrients, particularly Zn, will play an important role in stepping up the productivity of groundnut. In a field experiment on groundnut nutrition, the yield losses due to Zn deficiency were found to be 13.3% to 20% (Singh *et al*., 2004). More recently, substantial arable crop responses to Zn fertilization have been reported in Australia, India and Turkey, where wheat grain yields have increased by over 600% since the mid-1990s with the concomitant annual economic benefit of US $ 100 million (Cakmak, 2004). Particle size may affect agronomic effectiveness of Zn fertilizers. Decreased particle size also increases the specific surface area of a fertilizer, which should increase the dissolution rate of fertilizers with low solubility in water such as zinc oxide (ZnO) (Mortvedt, 1992). Granular zinc sulphate ($ZnSO_4$) (1.4 to 2 mm) was somewhat less fine $ZnSO_4$ (0.8 to 1.2 mm) whereas granular ZnO was completely ineffective (Allen and Terman, 1996). Gradual increase in Zn uptake could be observed with decreasing granule size and only the powder form produced plants with Zn concentrations in the sufficient range. Since granules of 1.5 mm weigh less than granules of 2.0 or 2.5 mm, smaller granules were used for the same weight, resulting in a better distribution of Zn, and the higher surface area of contact of Zn fertilizer resulted in better Zn uptake (Liscano et al., 2000). Therefore ample work has been done and emphasis was made o the particle size to increase the efficiency of the fertilizers for better uptake and higher yields.

Nanomaterials are proposed to be the materials for the new millennium. Carbon –based and metal based nanoparticles are most the commonly engineered and are often studied. Nanoparticles of size below 100 nm fall in the transition zone between individual molecules and the corresponding bulk materials, which generate both positive and negative biological effects in living cell (Nel *et al*., 2006). There is increasing amount of research on the biological effects of nanoparticles on higher plants. Several studies are concerned with the synthesis of nanomaterials using biological routes. Only limited studies have been reported on the promontory effects of nanoparticles on plants in low concentrations.

The present study was taken up to investigate the promontory inhibitory effects of various concentrations of ZnO nanoparticles on growth, development and final yield of groundnut (*Arachis hypogaea* L.) Nanoparticles with small size and large surface area are expected to be the ideal candidates for use as a Zn fertilizer in plants. Farmers are using both for soil and foliar applications, however the efficacy is low. Therefore, this study was initiated to generate new information on the efficacy of nanoscale zinc oxide on the growth and yield of groundnut. `

The major challenge for global food and nutrition security is to feed the increasing world population with nutritious food. Therefore in the future, it is essential to increase not only production but also of high quality food with the required level of nutrients and protein is the main challenge. Zinc is an essential micronutrient for humans, animals and plants. Indian soils are deficient in zinc and food crops grown on these soils and human beings living in this area are suffering from zinc deficiency. High altitude area of Visakhapatnam having more rainfall. Soil application of fertilizers lost due to heavy rains. Most of the nutrients leachate from soil resulting poor yields. To overcome this nutritional problem and crop production, conventionally bulk form of $ZnSO_4$ is applied to soil or to foliage as exogenous source, but most of the time the bulk forms are going to be fixed in the soil and making it not available to the rhizosphere and becomes toxic to the soil microorganisms and plants. Sustainable agriculture mainly aims to reduce application of chemical fertilizer and minimize nutrient losses in fertilization and increase yields through optimized nutrient management. Recently, nanotechnology is coming into focus because nano particles (NPs) are small in size (<100 nm) having high surface area and reactivity. Recent studies revealed that powder or Nano sized particles are found to be effective in absorption and translocation. But, physiological aspects of Nano zinc application and its accumulation in grains crops are meagre. Hence, the present study was carried out to investigate the effects of various concentrations of Zinc Oxide (ZnO) NPs on growth, yield and grain Zn content in groundnut.

MATERIALS AND METHODS

Characterization of ZnO Nanoparticles

ZnO nanoparticles of mean size of 25 nm diameter were used in the study. Nano crystal line zinc Oxide has been prepared by using the oxalate decomposition technique. Zinc oxalate was prepared by mixing equimolar (0.2 M) solution of zinc acetate and oxalic acid. The resultant precipitate was collected and rinsed extensively with double deionized water and dried in air. The oxalate was then ground and decomposed in air by placing it in a pre-heated furnace for 45 minutes at 500^0C. The characterization of the samples was done by transmission electron microscopy (HRTEM, JEOL 3010; Peabody, MA, USA), scanning electron microscopy (SEM FEI, Malvern, UK) and energy dispersive analysis of X-rays (EDAX, FEI Quanta200; FEI). The TEM samples were prepared by drop casting the suspension on carbon coated Cu grids.

Site of the field experiment

The field experiment was conducted during *Kharif* seasons 2018-19, 20219-20 2020-21, in Regional Agricultural Research Station, Chintapalle, ANGRAU. The experiment was laid down in randomized block design replicated three times. The gross plot size 8*6 m^2. Nine treatments T_1: control, T_2: RDF, T_3: RDF+ Soil application $ZnSO_4$@ 50 Kg/ha, T_4: RDF+ Foliar application $ZnSO_4$ 2 g/L at 25 & 45DAS, T_5: RDF+ Foliar application of nanoscale ZnO 150 ppm at 25 & 45 DAS, T_6: RDF+ Foliar application of nanoscale ZnO 200 ppm at 25 & 45 DAS, T_7: RDF+ Foliar application of nanoscale ZnO 400 ppm at 25 & 45 DAS, T_8: RDF+ Foliar application of nanoscale ZnO 50 ppm at 25 & 45 DAS, T_9: RDF+ Foliar application of nanoscale ZnO 100 ppm at 25 & 45 DAS were imposed.

The initial soil parameters were pH 6.1, Electrical conductivity 0.43 dS m^{-1}, Available Nitrogen 390 kg ha^{-1}, Available Phosphorus 18 kg ha^{-1}, Available potassium 379 kg ha^{-1}, Iron 7.86 ppm, Manganese 41.28, Zinc 0.26 and Copper 0.96 ppm. Soil texture was silt clay loam and Water holding capacity was 33%

RESULTS AND DISCUSSION

Table 1: Effect of Nano particulate zinc oxide on growth parameters (Pooled data)

Treatments	**Plant height (cm)**	**Root length (cm)**	**No of branches per plant**
T_1: control	26.46	8.04	4.2
T_2: RDF	35.50	9.80	5.1
T_3;RDF+ Soil application $ZnSO_4$@ 50 Kg/ha	39.00	10.80	6.6
T_4: RDF+ Foliar application $ZnSO_4$ 2 g/L at 25 & 45DAS	41.60	11.07	5.5
T_5: RDF+ Foliar application of nanoscale ZnO 150 ppm at 25 & 45 DAS	42.40	9.84	7.4
T_6: RDF+ Foliar application of nanoscale ZnO 200 ppm at 25 & 45 DAS	43.53	11.14	7.6
T_7: RDF+ Foliar application of nanoscale ZnO 400 ppm at 25 & 45 DAS	35.13	9.46	6.7
T_8: RDF+ Foliar application of nanoscale ZnO 50 ppm at 25 & 45 DAS	40.33	10.19	5.8
T_9: RDF+ Foliar application of nanoscale ZnO 100 ppm at 25 & 45 DAS	37.20	10.67	7.0
CD (0.05%)	7.48	1.73	1.41
SEm±	21.41	4.91	3.2

Growth parameters:

The plant height (43.53 cm) was recorded in RDF + Foliar application of nanoscale ZnO 200 ppm @ 25 and 45 Days after Sowing showed highest plant height compared to soil application of $ZnSO_4$. Highest root length was noted in treatment RDF along with Foliar application of nanoscale ZnO 200 ppm @ 25 and 45 Days after Sowing i.e 11.14 cm followed by RDF + Foliar application of $ZnSO_4$ 2g/L @ 25 and 45 Days after Sowing i.e 11.07 cm. The highest number of branches per plant was noticed in treatment with RDF+ Foliar application of nanoscale ZnO 200 ppm @ 25 and 45 Days after Sowing (7.6) compared to all other treatments. Similar results were observed by Prasad et al (2012), he suggested that ZnO NPs are absorbed by plants to a larger extent as compared to $ZnSO_4$ bulk. They also observed beneficial effects of NPs in enhancing plant growth, development and yield in peanut at lower doses, but at higher concentrations ZnO NPs were detrimental just as the bulk nutrients. Racuciu and Creanga (2007) when they analyzed the influence of magnetic nanoparticles coated with tetramethylammonium hydroxide on the growth of zea mays plants in early ontogenetic stages. Small concentrations of aqueous Ferro fluid added in culture medium had a stimulating effect on the growth of plantlets while higher concentrations of aqueous Ferro fluid induced an inhibitory effect. It was noted that water repellence potential of leaf surface acts as one of the limiting factors, which can affects the Zn uptake through spray application processes (Holder, 2007) The increase in vegetative growth in Groundnut (*Arachis hypogea* L) might be due to fundamental role Zinc in protecting and maintaining structured stability of cell membrane (Welch et al., 1982, Cakmak, 2000). Zn is used for protein synthesis, membrane function, cell elongation and tolerance to environmental stress (Cakmak, 2000). Plants emerging from seeds with low zn have poor seedling vigour and field establishment on zn- deficient soils.

Fig 1: Concentration of Zinc present in leaf and kernals

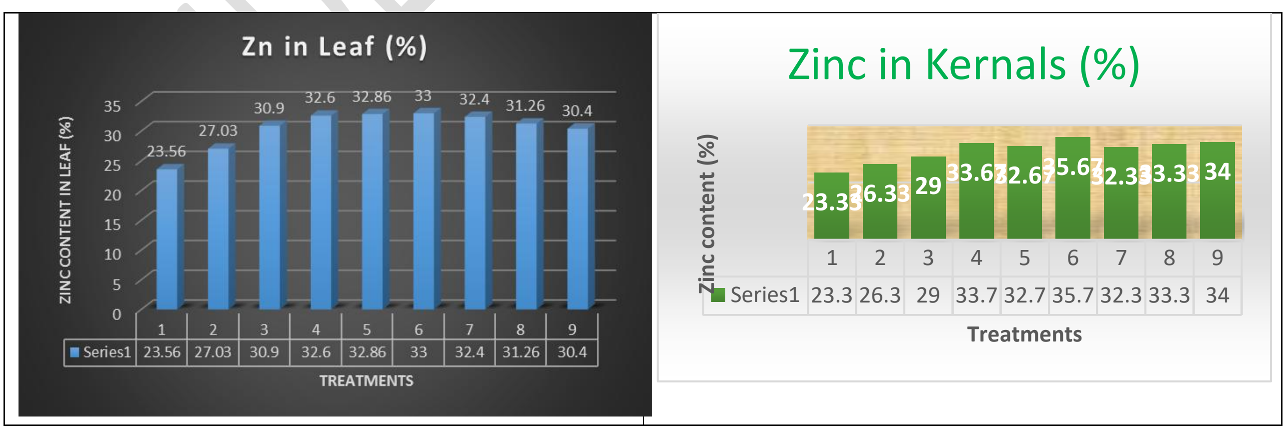

Table 2: Effect of Nano particulate zinc oxide on yield and yield attributes (Pooled data)

Treatment	No of pods per plant	no of unfilled pods	Shelling percentage
T_1: control	11.00	8.3	61.94
T_2: RDF	13.66	6.3	62.87
T_3;RDF+ Soil application $ZnSO_4$@ 50 Kg/ha	14.00	5.0	63.19
T_4: RDF+ Foliar application $ZnSO_4$ 2 g/L at 25 & 45DAS	16.00	6.3	64.22
T_5: RDF+ Foliar application of nanoscale ZnO 150 ppm at 25 & 45 DAS	17.00	5.0	70.77
T_6: RDF+ Foliar application of nanoscale ZnO 200 ppm at 25 & 45 DAS	18.00	5.3	72.48
T_7: RDF+ Foliar application of nanoscale ZnO 400 ppm at 25 & 45 DAS	15.66	7.3	63.46
T_8: RDF+ Foliar application of nanoscale ZnO 50 ppm at 25 & 45 DAS	15.00	5.6	72.30
T_9: RDF+ Foliar application of nanoscale ZnO 100 ppm at 25 & 45 DAS	16.66	5.6	69.10
CD (0.05%)	3.58	1.68	**7.96**
SEm±	9.80	5.89	22.9

Table 3. Effect of Nano particulate zinc oxide on yield and yield attributes (Pooled data)

Treatment	Pod yield ($Q\ ha^{-1}$)	Test weight (g)	100 Pod weight (g)
T_1: control	11.04	20.4	75.8
T_2: RDF	17.14	28.1	87.8
T_3;RDF+ Soil application $ZnSO_4$@ 50 Kg/ha	17.24	26.4	81.2
T_4: RDF+ Foliar application $ZnSO_4$ 2 g/L at 25 & 45DAS	20.66	22.4	92.8
T_5: RDF+ Foliar application of nanoscale ZnO 150 ppm at 25 & 45 DAS	23.75	24.9	98.7
T_6: RDF+ Foliar application of nanoscale ZnO 200 ppm at 25 & 45 DAS	24.36	30.9	96.7
T_7: RDF+ Foliar application of nanoscale ZnO 400 ppm at 25 & 45 DAS	17.91	27.9	96.0
T_8: RDF+ Foliar application of nanoscale ZnO 50 ppm at 25 & 45 DAS	19.08	23.2	94.9
T_9: RDF+ Foliar application of nanoscale ZnO 100 ppm at 25 & 45 DAS	19.19	28.6	96.6

CD (0.05%)	6.81	3.72	13.45
SEm(±)	18.60	9.54	32.35

Yield

The results revealed that pod yields of groundnut were greatly influenced by nanoscale zinc, increased grain yield upon application of RDF along with foliar application of Nanoscale ZnO 200 ppm at 25 & 45 DAS has showed highest pod yield (24.36 Q ha^{-1}) followed by treatment RDF+ Foliar application of nanoscale ZnO 150 ppm at 25 & 45 DAS i.e 23.75 Q ha^{-1} compared to all other treatments. RDF along with foliar application of ZnO 200 ppm at 25 & 45 DAS has showed highest test weight (30.9 g) which is on par with RDF. The highest 100 pod weight (98.7 g) was recorded with application of RDF+ Foliar application of nanoscale ZnO 150 ppm @ 25 and 45 Days after sowing. Which was on par with 200 ppm nano ZnO and RDF. Similar results reported by Prasad et al., (2012).

Foliar fertilization is more effective than soil application. Foliar zinc application significantly increased grain zinc concentration of groundnut indicating high mobility of zinc within plants. RDF + Foliar application of nanoscale ZnO 200 ppm at 25 & 45 DAS gave significantly higher peanut pod yield compared to no spraying. However soil application of ZnSO4 50 Kg per ha at sowing gave yields on par with no ZnSO4 application. This indicates that groundnut response to foliar application but not to soil application (Channabasava and Setty 1993). The effectiveness of various synthetic and natural chelates has been widely investigated (Alvarez and Gonzalez, 2006). A significant increase in number of pods per plant (14.97%), shelling percentage (3.56%) and pod yield (22%) due to the application of P and Zn were reported by Majumdar et al. (2001)

Table 4 : Effect of nanoscale Zinc Oxide on plant and kernel zinc

Treatment	**Zn (%) plants**	**Zn (%) kernels**
T_1: control	23.56	23.33

T_2: RDF	27.03	26.33
T_3;RDF+ Soil application $ZnSO_4$@ 50 Kg/ha	30.90	29.00
$T_{4:}$ RDF+ Foliar application $ZnSO_4$ 2 g/L at 25 & 45DAS	32.60	32.67
$T_{5:}$ RDF+ Foliar application of nanoscale ZnO 150 ppm at 25 & 45 DAS	32.86	33.67
$T_{6:}$ RDF+ Foliar application of nanoscale ZnO 200 ppm at 25 & 45 DAS	33.00	35.67
$T_{7:}$ RDF+ Foliar application of nanoscale ZnO 400 ppm at 25 & 45 DAS	32.40	32.33
$T_{8:}$ RDF+ Foliar application of nanoscale ZnO 50 ppm at 25 & 45 DAS	31.26	33.33
$T_{9:}$ RDF+ Foliar application of nanoscale ZnO 100 ppm at 25 & 45 DAS	30.40	34.00
CD (0.05%)	**2.62**	**7.24**
SEm±	7.43	21.5

Leaf and kernel samples

The post-harvest leaf and kernel sample analysis revealed a significant increment in zinc content in leaves and kernel. The highest zinc content in leaf (33.0%) was recorded with application of RDF along with foliar application of Nano ZnO 200 ppm at 25 & 45 DAS Compared to soil application of $ZnSO_{4.}$ The highest zinc content in kernel (35.6%) recorded with RDF along and foliar application of Nano ZnO 200 ppm at 25 & 45 DAS followed by the spraying of 100 ppm Nano scale ZnO @ 25 and 45 DAS and RDF compared to all other treatments.

The results suggested that the micronutrients, Zn can be delivered into peanuts seeds through ZnO nanoparticles. A higher amount of Zn was present in the seed when treated with nanoscale ZnO. This improves the germination, root growth, shoot growth and pod yield of the nano scale ZnO treated palnts.

The results point to the use of nanoscalematerials in agriculture, especially in peanut, one of the main source of livelihood in certain parts of the world. The results emphasized that nanoscale nutrients can be supplied to the crops either through seed dressing or by foliar application with much decreased doses to get the desired results.

Concentrated liquids suspensions of ZnO are used for foliar application but their performance is strongly determined by the size range specification of the ZnO particles present in the formulation (Moran, 2004). Leaf water repellency of adaxial or abaxial surface is a main limiting factor, which can affect the Zn uptake through spray application processes (Watanabe and Yamaguchi, 1991: Holder, 2007). The permeability of the cuticle to water and to lipophilic organic molecules increases with mobility (distribution co-efficient) and solubility (partition co-efficient) of these compounds within the transport-limiting barrier of the cuticles. Ions being highly water soluble might have some hindrance in penetrating the lipophilic cuticle. This may be act as limiting factor in Chelated $ZnSO_4$. But our custom-made nano particle ZnO, which is having less hydrophilicity and being more dispersible in lypophilic substances compared to the ions, can penetrate through the leaf surface (Da Silva *et al.*, 2006) compared to ZnSO4. Also the mobility of the nanoparticles is known to be very high which ensures the phloem transport and ensures the nutrient to reach all parts of the plant. The presence of Nano particles both in the extracellular space and within some cells in the living plant cubital pepo was reported (Gonzalez-Melendi et al., 2008). The bio availability of the nanoparticles because of its size and lower water solubility (which inhibit rapid falling off compared to ionic supplements) can also be highest compared to $ZnSO_4$. The inherent small size and the associated large surface area of nanoscale ZnO fertilizer may increase the uptake as reported earlier.

Regarding soil properties, the results revealed that there was no significance difference among the treatments regarding Avail. N, Available K and pH. The lowest Phosphorus (17.20 kg ha^{-1}) was recorded with RDF+ Foliar application of nanoscale ZnO 200ppm at 25 & 45 DAS and highest (24.46 kg ha^{-1})was recorded in T_9 (RDF+ Foliar application of nanoscale ZnO 100 ppm at 25 & 45 DAS) .

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