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Effect of Zinc Nano-Fertilizer on Growth and Yield of Wheat (*Triticum aestivum* L.) under Saline Irrigation Condition

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Abstract: Zinc deficiency in cereal crops can be solved by the application of Zn through bio fortification. Nanotechnology is one of the options to enhance the nutritional value of crops as some engineered nanoparticles (NPs) could be used as a fertilizer. Zinc can be used in the form of zinc oxide (ZnO) NPs. The present study used the direct precipitation method to prepare ZnO NPs, characterized it by using dynamic light scattering (DLS), scanning electron microscopy (SEM) and fourier-transform infrared spectroscopy (FTIR) and then evaluated the effect of Zinc nano-fertilizer on growth and yield of wheat (*Triticum aestivum* L.). The field experiment consisted of seed treatment, foliar application and combination of seed treatment as well as foliar application of bulk Zn sources and nano Zn source. Results showed that the seed treatment of ZnO NPs @1000 ppm followed by three foliar sprays of ZnO NPs @1000 ppm at 21, 45 and 90 days after sowing proved to be significantly superior in enhancing the plant height, number of effective tillers per plant, spike length, test weight, grain and straw yields, grain and straw zinc content and uptake by grain and straw.

Keywords: *Zinc oxide(ZnO), nanoparticles(NPs), use efficiency, nutrient uptake*

Introduction

Micronutrients are essential for increasing crop production and enhancing animal and human health. About one third of agricultural soils in the world are estimated to be low in available zinc (Zn), resulting in poor crop yields and nutritional quality of the harvested grains (Alloway 2008; Cakmak 2008). Among the micronutrients, Zn deficiency is the most detrimental to crop growth and yield of all the cereal crops including wheat (Alloway 2008; Marschner 1995). The deficiency of Zn in Indian as well as world soils is very well documented constraint in crop production and since last couple of decades, it is considered to be the 4th most yield limiting nutrient after N, P_2O_5, and K_2O in India (Katyal and Sharma 1991; Singh 2009; Shukla *et al*. 2014).

Cereals grains such as wheat (*Triticum aestivum* L.) contain only small concentrations of Zn. Wheat is the second most important crop in India after rice and contributes nearly 35 per cent to the national food basket. India will require 109 million tons of wheat to feed the population of 1.25 billion by 2020 A.D, which can be achieved by growth rate of 2.2 per cent but the current growth rate is only 1.0%.

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The viable options for ameliorating Zn deficiency in soils as well as to enhance Zn in major food crops is limited by poor fertilizer use efficiency, which must be improved. Nano-fertilizer is required in less quantity, so it reduces the cost of fertilizers and also reduces the chemical load of fertilizers in the soil. Nano-fertilizer facilitates slow and steady release of nutrients, thus enhances nutrient use efficiency by reducing loss of nutrients. The application of nano-technological formulation to agricultural crop inputs is one of the proposed tools for sustainable intensifications.

Nano-particles (NPs) could also be used as a source of essential plant nutrients (Rizwan *et al.* 2017; Moghaddasi *et al.* 2017).Among NPs, zinc oxide (ZnO) NPs are the most widely used NPs and their positive effects on the ecosystem have been reported (Rizwan *et al.* 2017).

Nano particles are expected to be the ideal candidates for use as a Zn fertilizer in wheat (Adhikari *et al.* 2015). The seed treatment of zinc nano-fertilizers, proved as growth booster in initial crop growth period, followed by foliar spray is effective in increasing growth of crops throughout their life period. Based on this background, the present experiment was carried out to evaluate the effects of seed treatment and foliar application of zinc nano-fertilizer on growth, yield, Zn content, uptake and zinc use efficiency by wheat.

Materials and Methods

Experimental details

The present experiment was carried out in two parts, (i) laboratory study for synthesis and characterization of zinc oxide nanoparticles and (ii) field study to evaluate the efficacy of synthesized ZnO NPs (zinc oxide nanoparticles) on growth and yield of wheat crop.

Laboratory study

The experiment was conducted in Department of Nanotechnology and Centre for Advanced Research in Plant Tissue Culture, Anand Agricultural University. ZnO NPs were prepared by adding 1 *N* NaOH(pH 10.5) to the 0.1 *M* $ZnSO_4.7H_2O$ solution. Then, it was centrifuged at 6000 rpm for 10 minutes. The collected pellet was washed with Milli Q DI-water and again centrifuged at 6000 rpm for 10 minutes. Further, 1000 ppm STPP (Sodium Tri polyphosphate) was added to the pellet and then the solution was sonicated for 10 minutes. Synthesized solution was dried at 100°C in hot air oven till the complete removal of water and thus dried powder obtained. After the preparation of ZnO NPs, different characterization techniques were used to investigate their particle size (nm), polydispersive index (PDI) and count rate (Kcps), zeta potential and morphological characteristics.

Field study

The present experiment was carried out at Agronomy Farm, College of Agriculture, Anand Agricultural University, Vaso (Gujarat) during rabi season of the year 2018-19.The soil had low available nitrogen, medium available phosphorus and available potash. The Zn was deficient but Mn, Fe and Cu were sufficient (Table 1).

Experiment consisting of ten treatments viz.T_1[control (only NPK)]; T_2[T_1 + seed treatment of ZnO NPs suspension at 1000 ppm]; T_3[T_1+seed treatment of ZnO (30% Zn) 10 mL per kg seeds]; T_4[T_1+foliar spray of ZnO NPs at 1000 ppm]; T_5[T_1+foliar spray of 0.5% $ZnSO_4$]; T_6[T_2+ foliar spray of ZnO NPs at 1000 ppm]; T_7[T_2+ foliar spray of 0.5% $ZnSO_4$]; T_8[T_3+ foliar spray of ZnO NPs at 1000 ppm]; T_9[T_3+ foliar spray of 0.5% $ZnSO_4$] and T_{10}[T_1+soil application of $ZnSO_4$ @25kgha^{-1} (as per STV)]was conducted in Randomized Block Design (RBD). Wheat variety GW 496 was raised under recommended dose of fertilizers (120 kg N and 60 Kg P_2O_5) was applied urea and diammonium phosphate, respectively. Phosphorus (60 kg P_2O_5ha^{-1}) and 50 per cent of nitrogen was applied as basal dose. Remaining 50 per centof nitrogen was applied two split applications at 21 and 35 days after sowing. Application of $ZnSO_4$ (25 kgha^{-1}) was carried out as per soil test value at the time of sowing.The seeds were soaked in ZnO NPs suspension of 1000 ppm for 2 hours and then shade-dried for an hour and then stored in plastic bags for sowing. The seeds (1Kg) were coated with slurry (10 ml) of ZnO (30% Zn).Three sprays of 1000 ppm ZnO NPs and 0.5% $ZnSOp_4$ were done at 21, 45 and 90 DAS in respective treatments.

Table 1.Properties of the experimental soil

Parameters	Value
Clay (%)	10.50
Silt (%)	10.20
Fine sand (%)	75.25
Coarse sand (%)	4.05
Texture	Sandy loam
pH (1:2.5)	8.75
EC (1:2.5) (dSm^{-1})	0.76
Organic carbon (%)	0.45
Available N ($kgha^{-1}$)	204
Available P_2O_5($kgha^{-1}$)	32.25
Available K_2O ($kgha^{-1}$)	206
Available S ($mgkg^{-1}$)	9.30
DTPA -Fe($mgkg^{-1}$)	5.23
DTPA -Mn($mgkg^{-1}$)	5.78
DTPA -Zn($mgkg^{-1}$)	0.40
DTPA -Cu($mgkg^{-1}$)	0.32

Growth and yield attributes

Days to emergence was recorded between 6 to 8 DAS.The plant height was recorded at 21, 45 DAS and at harvest. At maturity, average number of effective tillers per plant, spike length was also recorded from each plot. At harvest of the crop, grain and straw yield were computed and representative, grain and straw samples were collected for analysis. The samples were dried in paper bags at 70 ^{0}C till constant weight in a hot air oven and thereafter ground in a stainless steel grinder. Harvest index was calculated as follows:

Harvest Index (%) = [Economic yield ($kgha^{-1}$)/ Total biological yield ($kgha^{-1}$) ×100]

Soil, water and plant analysis

Soil samples were processed and analysed for pH and electrical conductivity (EC) (1:2.5) using glass electrode pH meter and EC meter, respectively. Organic carbon was estimated by Walkley and Black (1934) method, available N by alkaline potassium permanganate method (Subbiah and Asija 1956), available P by Olsen's method (Olsen *et al.* 1954). Available K by extraction with 1*N* ammonium acetate at pH 7.0 (Hanway and Heidal 1952) and available sulphur by turbidimetric method (Chesnin and Yien 1951). For analysis of available iron (Fe), copper (Cu), manganese (Mn) and zinc(Zn), soil samples were extracted by 0.005*M*DTPA containing 0.01*M* $CaCl_2$ and 0.1*M* TEA (pH-7.3) (Lindsay and Norvell 1978) and analysed by atomic absorption spectrophotometer. Finely ground straw and grain samples were digested with di-acid mixture (HNO_3:$HClO_4$: 3:1, v/v) and analysed for micronutrients using atomic absorption spectrophotometer. The underground (tube well) water samples were analysed for different water quality parameters following standard procedures..

Statistical analysis

The data were statistically analysed as per the methods by Steel and Torrie (1982). The value of 'F' was worked out and compared with value of 'F' at 5 per cent level of significance. The values of standard error (mean) (S.Em.±), critical difference (C. D.) and coefficient of variation (C.V. %) were also calculated and appropriately used for interpretation of data.

Results and Discussion

Effect of zinc nano-fertilizer on growth and yield attributes of wheat

At 21 DAS, maximum plant height (21.18 cm) was observed under T_6 (seed treatment of ZnO NPs suspension @1000 ppm) followed by three foliar sprays of ZnO NPs @1000 ppm at 21, 45 and 90 DAS, which was statistically at par with treatments T_2 and T_4.

At 45 DAS maximum plant height (63.78 cm) was observed under T_6 followed by three foliar sprays of ZnO NPs @1000 ppm at 21, 45 and 90 DAS which was statistically at par with treatments T_2 and T_4. Similarity at harvest, maximum plant height (93.60 cm) was observed under T_6, which was at par with T_2 and T_4 (Table 2).This increase in height was due to extended intermodal length. Such increase could be ascribed to higher precursor activity of ZnO NPs in auxin production (Kobayashi and Mizutani 1970). Adhikari *et al.* (2015) also reported that the application of ZnO nano particles on maize plant enhances plant growth as compared to conventional zinc fertilizer *i.e.* $ZnSO_4$.

The maximum number of tillers per plant (9.30) were recorded with T_6 followed by three foliar sprays of ZnO NPs @1000 ppm at 21, 45 and 90 DAS and was statistically at par withT_2 and T_4.

The maximum spike length (11.82 cm) was associated with T6 followed by three foliar sprays of ZnO NPs @1000 ppm at 21, 45 and 90 DAS) and was statistically at par with T2 and T4. The improvement in spike length under T6 was to the tune of 51.1 per cent over control (Table 2). The result is in agreement with the findings of Potarzycki and Grzebisz (2009).

Table 2. Effect of zinc nano-fertilizer application on growth attributes of wheat

Tr. No.	Treatment details	Days to emergence	Periodical Plant Height (cm)			No. of effective tillerplant^{-1}	Spike length (cm)
			21DAS	45 DAS	At Harvest		
1	Control	8.1	15.95	42.85	73.97	6.35	7.82
2	ST-ZnO NPs	7.6	20.94	59.98	88.00	9.20	10.78
3	ST-ZnO	8.0	17.89	51.10	74.94	6.90	8.63
4	FS-ZnO NPs	8.0	20.99	61.85	90.90	9.25	10.88
5	FS- $ZnSO_4$	8.0	17.67	52.65	75.52	6.95	8.86
6	ST-ZnO NPs+ FS-ZnO NPs	7.8	21.18	63.78	93.60	9.30	11.82
7	ST-ZnO NPs+ FS- $ZnSO_4$	7.9	18.68	57.35	79.51	7.10	8.83
8	ST-ZnO+ FS-ZnO NPs	8.0	19.23	55.11	81.83	7.73	9.95
9	ST-ZnO+ FS- $ZnSO_4$	8.0	18.21	53.30	74.98	7.30	9.10
10	SA- 25 kg/ha $ZnSO_4$ (STV)	8.0	17.75	50.35	73.75	6.80	8.47
	SEm ±	2.25	0.74	2.15	2.89	0.43	0.38
	CD at 5%	NS	2.15	6.23	8.38	1.26	1.09
	CV (%)	2.39	7.85	7.83	7.16	11.30	7.92

Effects of zinc nano-fertilizer on grain and straw yield of wheat

Seed treatment of ZnO NPs suspension at 1000 ppm followed by three foliar sprays of ZnO NPs at 1000 ppm at 21, 45 and 90 DAS recorded maximum test weight (49.28 g), which was at par with T_2 and T_4 (Table 3). These results are in conformity with the findings of Delfani *et al.* (2014), Laware and Raskar (2014) and Prasad *et al.* (2012).

The grain and straw yield of wheat were significantly improved due to different treatments. The grain yield (4984 kg ha^{-1}) was maximum with seed treatment of ZnO NPs suspension @1000 ppm followed by foliar sprays of ZnO NPs at 1000 ppm, which was at par with T2 and T4.Straw yield (6465kgha^{-1}) was maximum with seed treatment of ZnO NPs suspension @1000 ppm followed by foliar sprays of ZnO NPs at 1000 ppm, which was at par with T2 ,T3 and T4 (Table 3). The improvement in yield of grain and straw was to the tune of 21.4 and 33.4 per cent, respectively over control. Positive effect of Zn on grain yield in Zn deficient soil is one of the most widely documented facts (Patel 2011; Behera *et al*. 2015).

The effect of zinc nano-fertilizer application was not significant on harvest index of wheat over other the treatments and control.

Effects of zinc nano-fertilizer on zinc content and uptake in grain and straw of wheat

Seed treatment of ZnO NPs suspension @1000 ppm followed by foliar sprays of ZnO NPs @1000 ppm had maximum zinc content (31.70 and 85.65 mgkg^{-1}) in grain and stover of wheat (Table 4), however it was at par with T_4 in case of grain-Zn content and T_2, T_3, T_4, T_7, T_8 and T_9 in case of straw–Zn content.

The application of zinc nano-fertilizer significantly increased the wheat grain and straw zinc uptake. Seed treatment of ZnO NPs suspension @1000 ppm followed by foliar sprays of ZnO NPs @1000 ppm recorded maximum zinc uptake (157.93 and 553.81 gha^{-1}, respectively) in wheat grain and straw, however it was at par with T_4 in case of grain zinc uptake and T_2 and T_4 in case of straw zinc uptake.

The results are in corroboration with the findings of Prasad *et al.* (2012), Subbaiah *et al.* (2016)and Adhikari *et al.* (2016).

Table 3. Effect of zinc nano-fertilizer on yield attributes and yield of wheat

Tr. No.	Treatment details	Test weight (g)	Grain yield (kgha^{-1})	Stover yield (kgha^{-1})	Harvest Index (%)
1	Control	42.48	4106	4847	46
2	ST-ZnO NPs	48.17	4783	6217	43
3	ST-ZnO	44.63	4340	5652	43
4	FS-ZnO NPs	48.20	4831	6334	43
5	FS- $ZnSO_4$	45.13	4444	5499	45
6	ST-ZnO NPs+ FS -ZnO NPs	49.28	4984	6465	44
7	ST-ZnO NPs+ FS - $ZnSO_4$	45.18	4469	5443	45
8	ST-ZnO+ FS -ZnO NPs	46.54	4501	5580	45
9	ST-ZnO+ FS - $ZnSO_4$	45.48	4485	5427	45
10	SA- 25 kg/ha $ZnSO_4$(STV)	43.68	4436	5395	45
	SEm ±	0.46	161.8	300.5	1.4
	CD at 5%	1.34	469	872	NS
	CV (%)	2.01	7.13	10.57	6.45

Table 4. Effect of zinc nano-fertilizer on zinc content and uptake by wheat

Tr. No.	Treatment details	Zn content ($mgkg^{-1}$)		Zn uptake (gha^{-1})	
		Grain	Stover	Grain	Stover
1	Control	19.84	50.56	81.91	245.36
2	ST-ZnO NPs	24.03	79.85	114.81	496.75
3	ST-ZnO	21.00	75.15	91.31	424.46
4	FS-ZnO NPs	29.50	84.75	142.79	526.85
5	FS- $ZnSO_4$	25.98	64.55	115.40	356.50
6	ST-ZnO NPs+ FS-ZnO NPs	31.70	85.65	157.93	553.81
7	ST-ZnO NPs+ FS- $ZnSO_4$	28.20	74.05	126.05	402.04
8	ST-ZnO+ FS-ZnO NPs	28.60	77.20	128.35	431.24
9	ST-ZnO+ FS- $ZnSO_4$	27.30	75.70	122.14	411.03
10	SA- 25 kg/ha $ZnSO_4$ (STV)	21.15	52.45	93.95	279.80
	SEm ±	0.92	4.13	5.66	25.00
	CD at 5%	2.66	11.99	16.42	72.55
	CV (%)	7.13	11.48	9.63	12.11

Conclusion

The findings of the present study suggested that the wheat yield, zinc content in grain and straw could be increased with the soakingof seed with ZnO NPs suspension at 1000 ppm for 2 hours followed by three foliar sprays of ZnO NPs@1000 ppm at 21, 45 and 90 days after sowing.

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