Journal of Pharmacognosy and Phytochemistry
Available online at www.phytojournal.com

**E-ISSN:** 2278-4136
**P-ISSN:** 2349-8234
JPP 2018; 7(5): 2048-2052
Received: 28-07-2018
Accepted: 30-08-2018

**Pooja Solanki**
Department of Environmental Sciences, M.D. University, Rohtak, Haryana, India

**JS Laura**
Department of Environmental Sciences, M.D. University, Rohtak, Haryana, India

**Correspondence**
**JS Laura**
Department of Environmental Sciences, M.D. University, Rohtak, Haryana, India

# Effect of ZnO nanoparticles on seed germination and seedling growth in wheat (*Triticum aestivum*)

**Pooja Solanki and JS Laura**

**Abstract**
Zinc deficiency caused by inadequate dietary intake is a global nutritional problem, particularly in developing countries like India. Therefore, Zinc fortification of wheat and other cereal crops is being urgently required and highly prioritize in research. A seed germination and seedling growth was planned to evaluate the influence of zinc application in various forms as Nanoparticles and Bulk. Wheat seeds were separately treated with different concentrations of nanoscale zinc oxide (ZnO) and bulk zinc sulfate ($ZnSO_4$) and Zinc Oxide (ZnO) suspensions (a common zinc supplements), respectively and the effect this treatment had on seed germination, seedling vigor, Root and Shoot length, Fresh and Dry weight, chlorophyll content were studied. These particles proved effective in increasing shoot and root growth and chlorophyll.

**Keywords:** Nanoparticles, biofortification, micronutrient, zinc deficiency

## 1. Introduction

Zinc being an essential micronutrient for biological metabolism is receiving worldwide attention because of increasing reports of zinc deficiency in food crops as well as in humans [1, 2, 3]. It is required for the normal growth and development of both humans and plants [4]. It affects the multiple aspects of the immune system and is necessary for functioning of cell mediating immunity and natural killer cells in humans [5] (Prasad, 2008). In the same way, it is crucial for the enzyme functioning in plants. It is used in the gene expression, stress tolerance [6] and pollen formation [7].

Zinc deficiency is the most prevalent micronutrient deficiencies and severely affects one-third of the World's Population chiefly the rural communities [2, 8]. Inadequate intake of food having low zinc content is a major contributor to the prevalence of zinc deficiency in humans. Since wheat is the commonest cereal crop and contributed to the major calorie, protein and micronutrient inputs in the humans. In developing countries wheat provides over 50% of the daily calorific intake [3].

Monotonous diet especially on low zinc cereals like wheat is the main reason behind the zinc malnutrition in humans; moreover it has high phytate which further limits zinc bioavailability [9, 10]. Various reports are available indicating that 50% of the cereal growing soil around the World is deficient in zinc, which further lowers gain zinc content [1, 3] (Alloway, 2004; Cakmak, 2008). Moreover, processing of cereals after harvesting markedly decrease grain zinc and other micronutrients like iron [11, 12]. Hence there is a urgent need to increase the grain zinc content and bioavailability in the developing countries [9, 13] (Welch and Graham, 2004; Zhao and McGrath, 2009).

In response to the aforesaid problem, different approaches have been suggested and applied in developing nations [14, 15]. The Biofortification is receiving great attention [16, 17] now a days to overcome the problem of micronutrient deficiency. Using conventional fertilizers for the fortification purpose have led to increased soil toxicity, so there is need to look for novel and smart nutrient delivery systems. Nanoparticles with smaller size, high surface are and high dissolution rates are the solution to that. Studies have reported about having positive effect of ZnO Nanoparticles on growth of *Vigna radiata*, *Cicer arientinum*, Cucumber, alfalfa, tomato etc. The effect of using nanoparticles as fertilizers varies from species to species. A very few studies are available on the effect of ZnO nanoparticles on crops especially on Wheat, which is the most common staple food for most of the developing country population. The aim of this study was to investigate the effects of different concentration of ZnO nanoparticles on seed germination, and seedling growth characteristics in Wheat (*Triticum aestivum)*.

## 2. Materials and Methods

### 2.1 Synthesis of Zinc Oxide Nanoparticles and seed treatment

Zinc oxide nanoparticles were prepared using simple chemical methods using Zinc Sulfate (0.1M) and Sodium Hydroxide (0.2M) in methanol according to [18]. Nanoparticles of 40-50 nm having a UV-maxima peak at 350nm were formed. The synthesized ZnO-NPs (40-50 nm) were suspended in water and used for treatment. Wheat (*Triticum aestivum*) seeds of variety WH-711 were procured from the CCSHAU, Regional Centre, Rohtak. The healthy seeds were selected to minimize error in seed germination. Seeds were surface sterilized with 2% Sodium Hypochlorite and rinsed twice with distilled water. The experimental treatments included five concentrations (250, 500, 1000, 1500 PPM) of ZnO NPs as well as the conventionally used fertilizers like $ZnSO_4$ and ZnO along with Control.

**2.2 Seed Germination:** 10 Seeds of equal size were placed in the perti-dishes (100mm×15mm) lined with 2 layers of sterilized filter paper. 10 ml of corresponding solution/suspension was added to the petri dishes and were placed in incubator at 26±1° C for 12 days. The percent germination was recorded 5 days after incubation. After 12 days the pertidishes were removed and data was recorded for root length, shoot length, fresh weight and dry weight. Seedling vigour index (SVI) was calculated as per the formula

$$SVI = \text{Germination } \% \times (\text{Root length} + \text{Shoot Length})$$

**2.3 Chlorophyll:** Total Chlorophyll was determined by the method of [19]. 50mg of fresh leaf material was hand homogenized in 10ml of 80% acetone and the absorbance was recorded at 645 and 663nm using spectrophotometer (Hitachi Model U-2000).

Chlorophyll content was calculated using following formula-

$$\text{Chlorophyll a (mg/g)} = 12.7\ (A_{663} - 2.69(A_{645}) \times \frac{V}{1000 \times W}$$

$$\text{Chlorophyll b (mg/g)} = 22.9\ (A_{645}) - 4.68(A_{663}) \times \frac{V}{1000 \times W}$$

$$\text{Total Chlorophyll} = 20.9 \times (A_{645}) - 8.02 \times (A_{663}) \times \frac{V}{1000 \times W}$$

Where, A=Absorbance, V= Final volume of sample made, W=Fresh weight of sample taken.

## 3. Result and Discussion

### 3.1 Seed germination

Highest germination percentage of 99% was recorded with Zinc oxide NPs at a concentration of 500 PPM. And the minimum germination percentage was observed with control. It has been reported by several workers that seed treatment with Zn induces a range of biochemical changes in seed, required to start the germination process, such as breaking of dormancy, hydrolysis or metabolization of inhibitors, imbibition and enzyme activation [20, 21]. It was also observed that the ZnO and ZnSO4 Bulk have a lower efficacy in improving the seed germination as compared to the ZnO NPs. These results are in agreement with [22] where the percent germination increase has been reported with the application of ZnO NPs on *Cicer arietinum*.

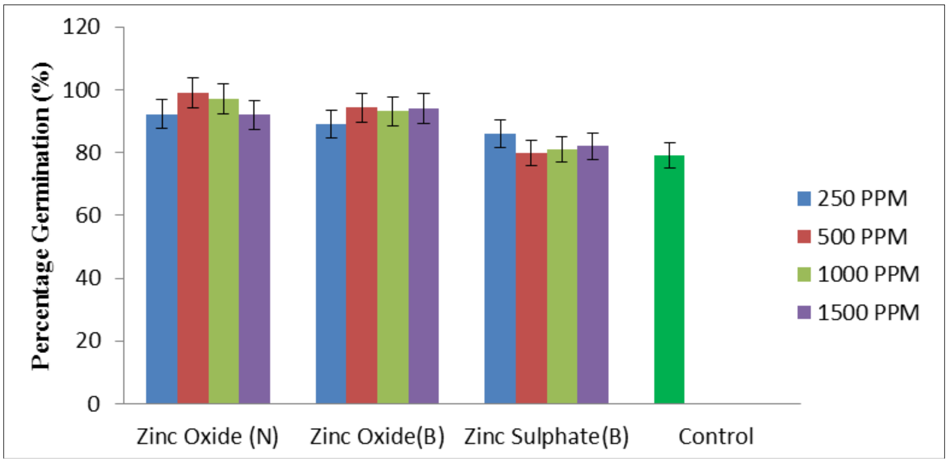


**Fig 1:** Germination percentage of wheat seeds affected by different Zinc Treatments along with Control


**Plate 1:** Germination of wheat seeds under different treatments

### 3.2 Seedling growth
### 3.2.1 Shoot and Root Length

The highest shoot Length of 13.6 cm was observed with the application of ZnO NPs at a concentration of 500 PPM and the lowest of 6.1 cm was observed with the control. A decrease in the shoot length has been observed with further increasing the concentration of zinc with an exception in case of ZnO NPs at 1500 PPM. It has been observed that there is a percentage increase of 122%, 90% and 59% over control at a concentration of 500 PPM for ZnO NPs, ZnO Bulk and ZnSO4 respectively in shoot length. Moreover, ZnO NPs is 17% more efficient than ZnO Bulk and ZnO Bulk is 40% more efficient than ZnSO4.

The highest root length was observed with ZnO NPs at an concentration of 500 PPM i.e. 7.9 cm and lowest is recorded with the control. There is an increase of 216, 164 and 144 % over control in ZnO NPs (500PPM), ZnO Bulk (250PPM) and $ZnSO_4$ (250 PPM) respectively. Moreover an increase of 19.6% in ZnO NPs (500 PPM) over Bulk ZnO (250 PPM) and 7.5% in ZnO Bulk (250 PPM) over $ZnSO_4$ bulk has been observed. It is noteworthy, that higher concentration of Zn causes significant reduction in root as well as shoots length. Similar findings were reported in groundnut where ZnO NPs at lower concentration increased the root and shoot length [23]. Such increase in the growth at lower concentration could be ascribed to higher precursor activity of Zn, especially ZnO NPs in auxin production [24].

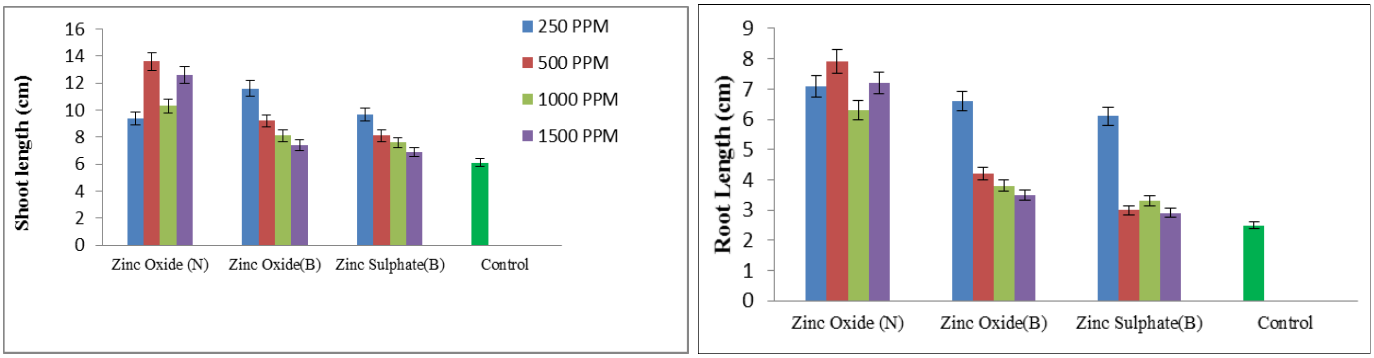


**Fig 2:** Shoot and root length of wheat seeds affected by different Zinc Treatments along with Control


**Plate 2:** Effect of different Zinc treatments on Wheat seedling after 12 days of incubation

**3.2.2 Fresh and Dry weight** Nano-ZnO (500PPM) has the highest fresh and dry weight of 0.34g and 0.048 g respectively. However the lowest fresh as well as the dry weight was observed to be of control i.e. 0.08g and 0.004g. The fresh weight of the seedling decreases with increasing concentration of ZnO NPs. The fresh of ZnO NPs (500PPM) treated seeds was 78.9% higher than the bulk ZnO (250 PPM) and 112.5% higher than the ZnSO4 (250PPM). Similarly, the dry weight of ZnO NPs (500PPM) treated seeds was 65% higher than the ZnO Bulk (250PPM) and 152% higher than the $ZnSO_4$ (250PPM) treated seeds. Similar results have found by the [25] in *Vigna radiata* in foliar spray of zinc oxide nanoparticles.

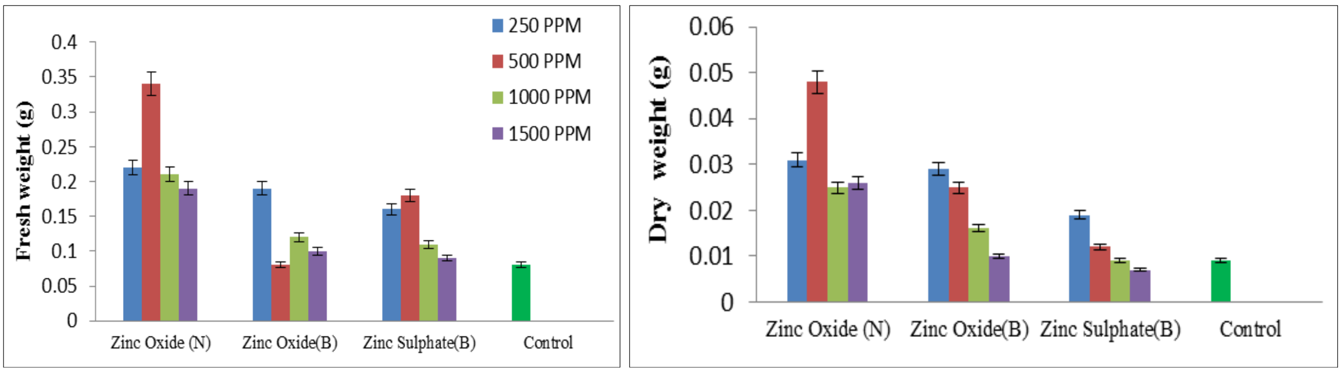


**Fig 3:** Fresh and Dry weight of wheat seeds affected by different Zinc Treatments along with Control

### 3.3 Total Chlorophyll Content

Total Chlorophyll content was found to be highest in ZnO NPs at an concentration of 500 PPM and the lowest in control. The Total chlorophyll content was found to be 280% higher than the control in ZnO NPs (500PPM), 80% in ZnO Bulk (250 PPM) and 66% higher in case of $ZnSO_4$ Bulk (250 PPM). It has been reported that Zn is an important factor in Chlorophyll synthesis [3].

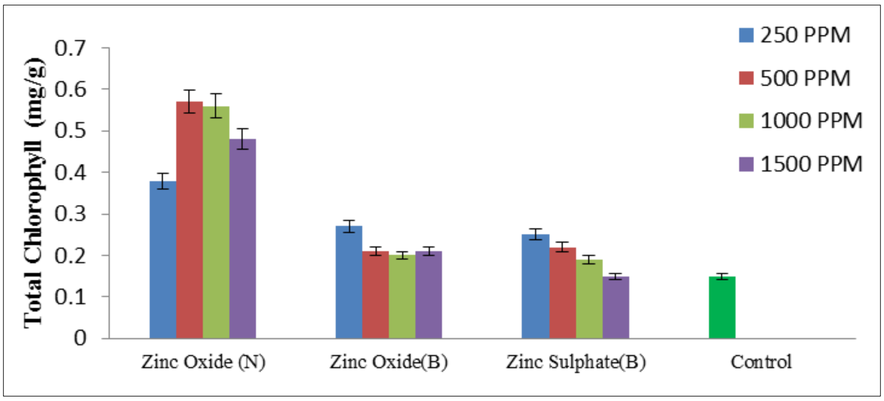


**Fig 4:** Total Chlorophyll of wheat seeds affected by different Zinc Treatments along with Control

### 3.4 Seedling Vigour Index

In general, all the ZnO treatments were found significantly superior over control with no zinc. Among different Zn treatments, ZnO NPs at 500 PPM recorded the highest seedling Vigour Index of germinated seeds. Though, different levels of bulk ZnO and $ZnSO_4$ has enhanced seedling vigour index but the magnitude of increase was less than their corresponding nano levels.

Plants emerging from low Zinc concentration have poor seedling vigour [26] and it has been reported that increasing seed Zn content significantly improves root and shoot growth of seeds under Zn deficiency. They have opinioned that high Zn concentration in seed could act as a starter fertilizer.

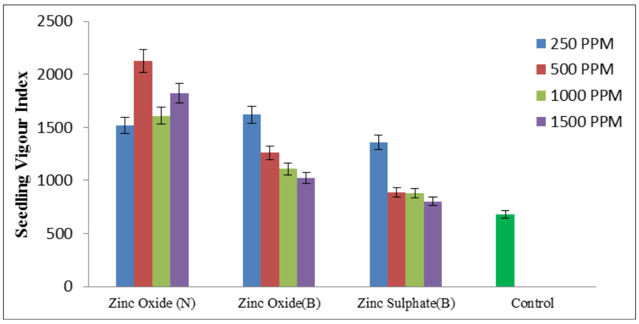


**Fig 5:** Seedling Vigour Index of wheat seeds affected by different Zinc Treatments along with Control

## 4. Conclusion

In the present study we have tried to evaluate the effect of ZnO NPs on the wheat Germination and seedling growth. NPs of 40-50nm were used along with the Bulk ZnO and $ZnSO_4$ at different concentration. The Zinc in any of the three mentioned form have shown positive effect on seedling growth over control. The NPs at a concentration of 500PPM have highest positive effect in terms of enhanced seed germination, root growth, shoot growth, Total Chlorophyll and seedling vigour Index. However, the growth decreased with increased concentration of Zn in the treatment. The ZnO NPs at every concentration are better than their Bulk counterparts in increasing the seedling growth. So, applying Zn in Nanoparticles form may help to increase the crop yield.

## 5. References


1. Alloway BJ. Zinc in Soils and Crop Nutrition. Brussels: IZA Publications, 2004.
2. Hotz C, Brown KH. Assessment of the risk of zinc deficiency in populations and options for its control. Food Nutrition Bulletin. 2004; (25):S91-S204.
3. Cakmak I. Enrichment of cereal grains with zinc: agronomic or genetic bio fortification? Plant Soil. 2008; 302:1-17.
4. Hafeez B, Khanif M and Saleem M. Role of zinc in plant nutrition: a review. American Journal of Experimental Agriculture. 2013; 3:374-391.
5. Prasad AS. Clinical, anti inflammatory and antioxidant role of zinc. Experimental Gerintology. 2008; 43:370-377.
6. Cakmak I. Role of zinc in protecting plant cells from reactive oxygen species. New Phytology. 2000; 146:185-205.
7. Pandey N, Pathak GC, Sharma CP. Zinc is critically required for pollen function and fertilization in lentil. Journal of Trace Elemental and Medical Biology. 2006; 20:89-96.
8. Stein AJ. Global impacts of human mineral malnutrition. Plant Soil. 2010; 335:133-154.
9. Welch RM, Graham RD. Breeding for micronutrients in staple food crops from a human nutrition perspective. Journal of Experimental Botany. 2004; 55:353-364.
10. Cakmak I, Kalayci M, Kaya Y, Torun AA, Aydin N, Wang Y. Biofortification and localization of zinc in wheat grain. J Agric. Food Chem. 2010a; 58:9092-9102.
11. Zhang Y, Song Q, Yan J, Tang J, Zhao R and Zhang Y. Mineral element concentrations in grains of Chinese wheat cultivars. Euphytica. 2010b; 174:303-313.
12. Kutman UB, Yildiz B, Cakmak I. Improved nitrogen status enhances zinc and iron concentrations both in the whole grain and the endosperm fraction of wheat. Journal of Cereal Science. 2011; 53:118-125.
13. Zhao FJ, McGrath SP. Bio fortification and phytoremediation. Current Opinion Plant Biology 2009; 12:373-380.
14. Bouis HE. Micronutrient fortification of plants through plant breeding: can it improve nutrition in man at low cost? Proc. Nutrition Society. 2003; 62:403-411.
15. Pfeiffer WH and Mc Clafferty B. Harvest Plus: breeding crops for better nutrition. Crop Science. 2007; 47:S88-S105.
16. Zhao FJ, McGrath SP. Bio fortification and phytoremediation. Current Opinion Plant Biology. 2009; 12:373-380.
17. Bouis HE, Welch RM. Biofortification-a sustainable agricultural strategy for reducing micronutrient malnutrition in the global south. Crop Science. 2010; 50:S20-S32.
18. Rodrigues-Paez J, Caballero AC, Villegas M, Moure C, Duran P, Fernandz JF. Controlled precipitation methods:

formation mechanism of ZnO nanoparticles. Journal of. European Ceram Society. 2001; 21:925-930.

19. Aron D. Copper enzymes isolated chloroplasts, polyphenol oxidase in Beta vulgaris. Plant Physiology. 1949; 24:1-15.
20. Harris D, Rashid A, Miraj G, Arif M, Shah H. On-farm seed priming with zinc sulphate solution - A cost-effective way to increase the maize yields of resource-poor farmers. Field Crops Research. 2007; 10:119-127.
21. Samad A, Khan MJ, Shah Z, Jan MT. Determination of optimal duration and concentration of zinc and phosphorus for priming wheat seed. Sarhad Journal of Agriculture. 2014; 30(1):27-34.
22. Avinash CP, Sharda SS, Raghvendra SY. Application of ZnO nanoparticles in influencing the growth rate of *Cicer arietinum*. Journal of Experimental Nano Science. 2010; 5(6):1-12.
23. Prasad TNVKV, Sudhakar P, Sreenivasulu Y, Latha P, Munaswamy V, Raja Reddy K, Sreeprasad TS, Sajanlal PR and Pradeep T. Effect of nanoscale zinc oxide particles on the germination, growth and yield of peanut. Journal of Plant Nutrition. 2012; 35:905-927.
24. Kobayashi Y, Mizutani S. Studies on the wilting treatment of corn plant: The influence of the artificial auxin control in nodes on the behavior of rooting. Proceedings of the Crop Science Society of Japan. 2013; 39:213-220.
25. Dhoke SK, Mahajan P, Kamble R, Khanna A. Effect of nanoparticles suspensions on the growth of mung (*Vigna radiata*) seedlings by foliar spray method. Nanotechnology Development. 2013; 3(1):1-5.
26. Yilmaz A, Ekiz H, Torun B, Gultekin I, Karanlik S, Bagci SA *et al*. Effect of different zinc application methods on grain yield and zinc concentration in wheat grown on zinc-deficient calcareous soils in Central Anatolia. Journal of Plant Nutrition. 1987; 20:461-471.