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Effect of zinc oxide nanoparticles on Germination and Growth Characteristics in Wheat Plants (*Triticum aestivum L.*)

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Abstract — *Nanotechnology opens a large scope of novel applications in biotechnology and agricultural industries. Despite the plenty of information available on the toxicity of nanoparticles to plant system, few studies have been conducted on mechanisms, by which nanoparticles exert their effect on plant growth and development. The present study was aimed to investigate the effects of zinc oxide nanoparticles on wheat (*Triticumaestivum *L.) plants. Zinc oxide nanoparticles has been synthesized by the simple chemical route. Synthesized nanoparticles have been confirmed with X-ray diffractometer (XRD), Transmission Electron Microscopy (TEM). Different concentration of synthesized zinc oxide nanoparticles (2000-10,000 ppm) solution were prepared and applied to healthy wheat seeds (variety: lok-1) by soaking approach. The seed germination, seedling vigor, plant growth characteristics were measured by using standard biophysical techniques and studied. Results of the present study showed enhancement in germination and growth characteristics in 5 days grown wheat seedlings for control upto 1000 ppm. However, above 1000 ppm, the significant decrease was observed in these parameters upto 2000 ppm. In addition, shoot length, seedling length, and root dry matters were affected by nanosized ZnO (~3-5 nm mean particle size) concentrations, significantly. These results indicate that employing nanosized ZnO in suitable concentration could endorse the seed germination of wheat in comparison to untreated control. Otherwise high concentrations had inhibitory or any effects on wheat.*

Keywords- *Wheat (Triticum aestivum L.), Seedling, germination, Zinc oxide nanoparticles*

I. INTRODUCTION

Nanotechnology, an emerging technology has been found to solve many of the agricultural-related problems with remarkable improvement. Nanotechnology has the potential to protect plants, monitor plant growth, detect plant and animal diseases, increase global food production, enhance food quality, and reduce waste for "sustainable intensification". Food and agricultural production are among the most important fields of nanotechnology application[1]. Also, it has the potential to revolutionize the agriculture with new tools to enhancing the ability of plants to absorb nutrients. The environmental impacts of these materials are rarely studied and reported in literature. A number of researchers have reported the essentiality and role of Zinc for plant growth and yield[2-6]. Zinc is required for chlorophyll production, pollen function, fertilization and germination [7-9]. Some Plant species, i.e. rape, corn,corn, lettuce, radish, ryegrass, cucumber[10] and wheat [11] are sensitive toward ZnO NPs. Presence of ZnO NPs in surrounding environment affect plant architecture, physiology and biochemistry. The toxicity is considered due to internalization of NPs, accumulation in root tissue and root surface, dissolution of zinc ions from NPs along with other physio chemical properties[12]. In many studies, there was increasing evidence in growth parameters of plants. Different concentrations such as 500 ppm and 1000 ppm of ZnO NPs in soyabean, Ramesh et .al[13]; 1.5 ppm in chickpea, reported that lower concentrations of concentrations of ZnO NPs exhibited beneficial effect on seed germination. ZnO Nanoparticles caused concentration dependent inhibition of root length when treated with ZnO NPs; the root growth of Garlic was completely blocked[14]. The properties of ZnO Nanoparticles (400 ppm and 1000 ppm) are responsible to give higher yield in peanut compare to chelated $ZnSO_4$ [15]. Laware and Raskar[16] studied the effects of different ZnO Nanoparticles (10 ppm to 40 ppm) on seed germination and seedling growth in Onion observed that seed germination increased in lower concentrations of ZnO Nanoparticles but showed decreased in values at higher concentrations. Yilmaz [17] studied the effects of zinc content on grain yield and zinc concentrations of wheat growth in zinc deficient calcareous soils. They observed that wheat plants emerging from seeds with low Zn content had poor seedling vigor and field establishment on zinc deficient soil. ZnO NPs induced a significant improvement in *Cyamopsistetragonoloba* plant biomass, shoot and root growth, chlorophyll and protein synthesis, acid and alkaline phosphatase in cluster bean rhizosphere [18]. Also, ZnO NPs promoted the root and shootlengths and root and shoot biomass in *Vignaradiata*and*Cicerarietinum* [19]. Rosa et al. [20] (1600 ppm ZnO NPs and 250 ppm of Zn^{2+}) applied different concentrations of ZnO NPs on cucumber, alfalfa and tomato, and found that only cucumber seed germination was enhanced. Thus, the effect of NPs on germination depends on concentrations of NPs and may vary from plants to plants. A very few studies have been done on the effects of high concentration of nanoparticles on crops particularly on wheat which is one of the most important crops in the world. The aims of this study were to investigate the effects of different concentrations of ZnO nanoparticles on seed germination, growth characteristics in Wheat *(Triticum aestivum).*

II. MATERIALS AND METHODS

ZnO nanoparticles were prepared using simple chemical route. Zinc oxide (ZnO) NPs about ~3-5 nm sizes were synthesized by dissolving 20 ml of zinc chloride (0.1M) ,100 ml of sodium hydroxide (0.1 M NaOH) and thioglycerol (0.1 M TG).in methanol.[21]. The synthesized ZnO-TG NPs were dried in oven, Suspended in water and then used for treatment.Wheat (*Tritium Aestivum.*) seeds of variety Lok-1 were procured from "Sheti Udyog Bhandar" Pune, Maharashtra, India. The healthy seeds with uniform size were selected to minimize errors in seed germination and seedling vigor. The seeds were surface sterilized in 5% sodium hypochloride solution for 15 min and then rinsed with distilled water for several times to remove excess of chemical. In order to study the effect of different concentrations of nanosized ZnO on wheat germination, a randomized completely design with four replications was employed. The experimental treatments included five concentrations (2000, 4000, 6000, 8000 and 10000 ppm) of nanosized ZnO and untreated control. To avoid aggregation of NPs, the medium was sonicated for 30 min. The experiment was conducted in laboratory conditions with natural light and an average temperature of 20±1°C.One hundred twenty seeds of similar size were randomly selected and placed on moistened paper as five groups of 20 seeds in beakers, and then 10 ml of each concentration treatment was added to each beaker. For the control, only distilled water was added to beaker. Seeds soaked in distilled water were acted as control. All these 6 beakers were kept in dark and after 24 hours seeds were taken out for plantation. Soaked seeds were planted on 0.8 % agar gel. In this study, agar gel used as growing medium or a substrate to reduce the errors as soil contains various types of salts and nutrients. 12 healthy seeds were selected from 20 soaked seeds and these seeds were then sowed in agar gel equidistantly in circular manner. These beakers were properly wrapped with polythene bags and kept under ambient conditions. Every day, the number of germinated seeds was recorded. Seeds were regularly watered with distilled water. The same measurement was taken after every 24 hours upto 5 days A seed was considered germinated when the radical was visible. At the end of experiment plants were isolated and analyzed for shoot, root length, and fresh weight (FW). The numbers of seeds germinated of each sample after 24 hours of sowing were measured.

In this study, we used following germination parameters: Weighted germination index (WGI), Final percentage germination (GP) for each treatment was calculated after seven days. These parameters were also calculated by using formula,

% germination = (no. of seeds germinated/total no. of seeds taken) x100 (1)

Also, vigor index for each sample was calculated by using the following formula, Seedling vigor was computed based on the formula [22]

Vigor index = % germination (Mean shoot length + Mean root length) (2)

After an incubation period of days, plumule and radical length of seedlings were measured using a ruler.

III. Results & Discussions

The results for the ZnO nanoparticles followed by a mechanistic evaluation of the various pathways are reported in the following sections.

3.1 Characterization of ZnO nanoparticles

The size and morphology of the ZnO nanoparticles were investigated using TEM. Fig.1 shows an TEM image of the nanoparticle sample.

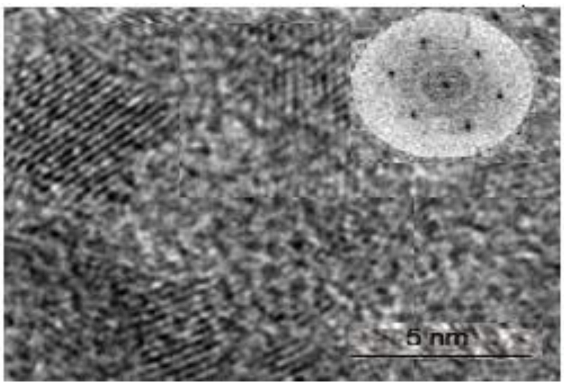

Fig.1 High resolution transmission electron micrograph of ZnO nanoparticles

It shows the presence of spherical monodispersed particles and the lattice of ZnO is clearly seen. XRD analyses (Fig 2) revealed single phase ZnO Wurtzite crystal structure. Particles with ~3-5 nm size can be seen. Particle size calculated from the XRD and that from the TEM is comparable to each other.

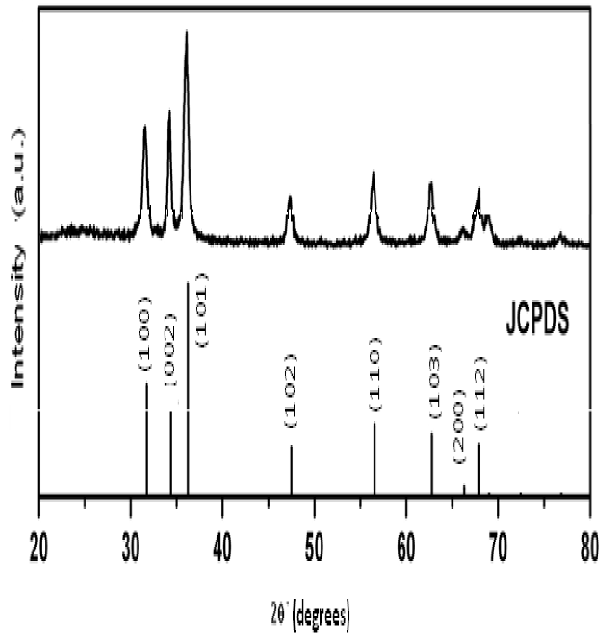

Fig.2 X-ray diffraction Pattern of ZnO nanoparticles

3.2 Seed Germination

In this study, we assessed the impact of ZnO nanoparticles on the most sensitive stages of plant development, i.e. seed germination and seedling growth. Among wheat germination indices, seed germination rate was affected by applied treatments. The percentage seed germination of nanoparticles treated wheat seedlings was reduced as compared to control sample

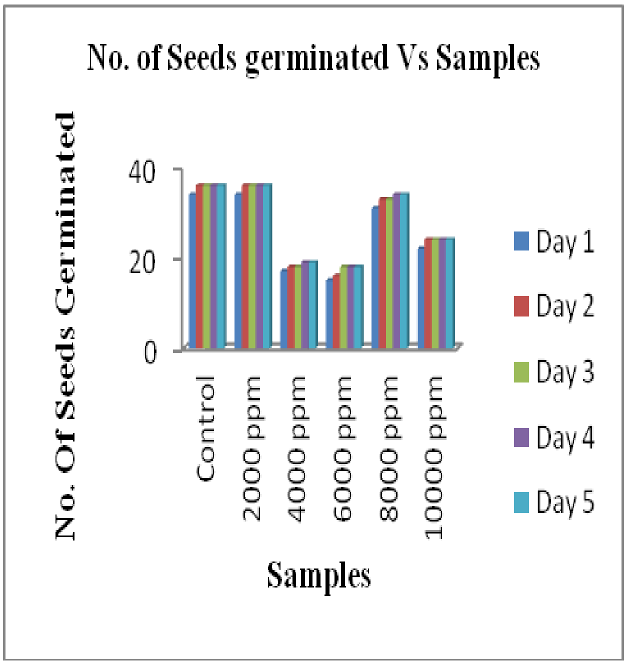

Fig.3. Average Seed Germination in control and Nanoparticles treated seeds

In control, the seed germination was 100 %. Same percentage seed germination was observed in 2000 ppm sample. However, after that percentage seed germination was decreased as concentration of nanoparticles increased (figure3,4).

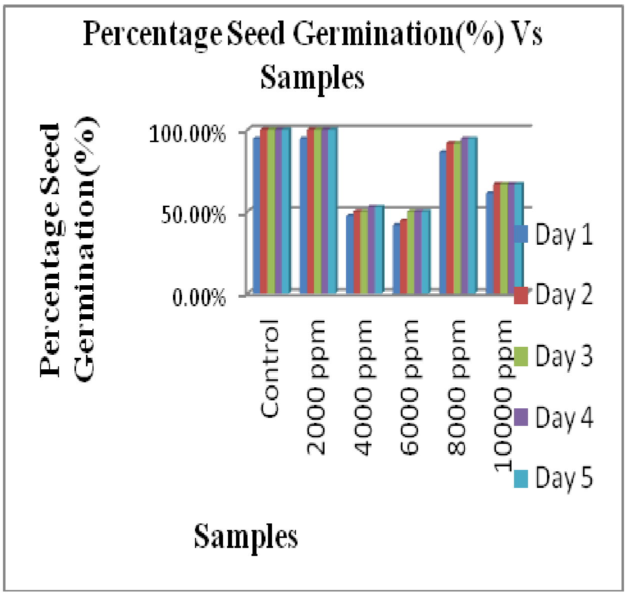

Fig 4. Percentage Seed Germination

3.2 Growth and Vigor Index

The growth of 5 days old control and nanoparticles treated seedlings is shown in figure 5. Results showed that the growth of nanoparticle treated seedlings reduced with increase in concentration except slight increase in growth at 8000 ppm. As shown in figure 6 the shoot length and root lengths are decreased with increase in concentrations of nanoparticles. However, no significant change was observed in number of secondary roots. Similar trend was observed for shoot weight and root weights . Vigor Index calculated from percentage seed germination and plant indicates the capacity for survival or strong healthy growth in plant. Results of the present study showed decrease in vigour index in nanoparticles treated seedlings compared control(figure 7).

Fig.5. Photographs of five days old control, and Nanoparticles treated wheat seedlings

As nanoparticle concentration increases as vigour index decreases except 8000 ppm showed the maximum seedling vigor index as compared to1000 ppm. Such inhibitory effects of nanoparticles were also reported by Lin and XING[10].on radish, rape and rye grass.

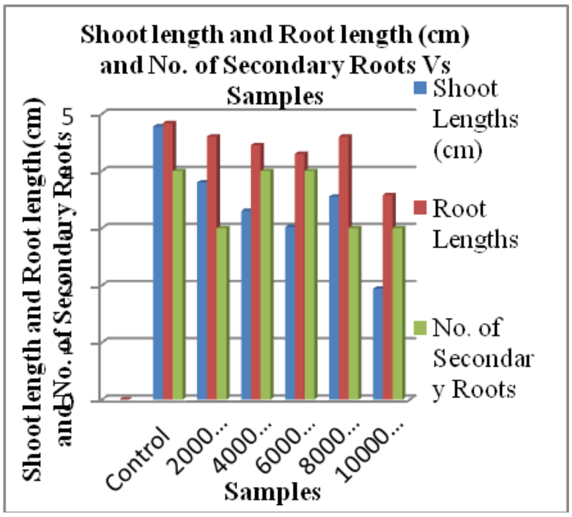

Fig 6. Average Shoot and Root Lengths and No. of Secondary Roots

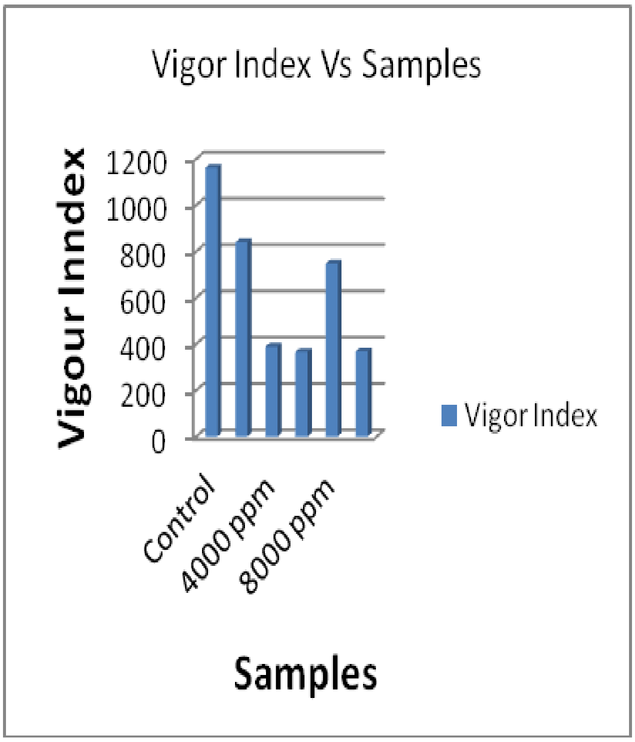

Fig 7. Vigor Index Vs Samples

IV. CONCLUSION

In summary, we have experimentally demonstrated the effect of ZnO NPs concentration on the wheat seed germination , the seedling vigor and plant growth characteristics. Results of the present study showed enhancement in germination, growth in 5 days grown wheat seedlings for control upto 1000 ppm. However, above 1000 ppm, the significant decrease was observed in these parameters upto 2000 ppm. In addition, shoot length, seedling length, and root dry matters were affected by nanosized ZnO concentrations. These results indicate that employing nanosized ZnO in suitable concentration could endorse the seed germination of wheat in comparison to untreated control. Otherwise high concentrations had inhibitory or any effects on wheat. The results of this study imply that further investigations should be made to determine impacts of zinc oxide NPs on other agricultural crops as well.

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