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Effect of Nano ZnO Seed Priming on Rice

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***Abstract* – Zinc is an essential element for crop growth, animal health, and human life. It is a vital trace element and acts as a cofactor or activator of many enzymes involved in various metabolisms. More than 50 percent of Indian soils are deficient in Zinc. Zinc enrichment through seed priming and fertilization has become a method of choice to increase grain zinc and yield. ZnO nanoparticles were synthesized by direct precipitation method using 1M zinc acetate and 2M sodium hydroxide as precursors. The synthesized nanoparticles were characterized using Fourier Transform Infrared Spectroscopy (FTIR). For seed priming various concentrations of ZnO nanoparticles ranging from 200–2000 ppm were used with four rice varieties viz., ADT-50, ADT-45, ADT-43, and CR-1009. The results of the seed priming experiment indicated the significant influence of Zn oxide nanoparticles @1000 ppm in increasing the shoot length, root length, vigour index, and germination percentage of various rice varieties. The 1000 ppm ZnO seed priming recorded the highest shoot length (12.94 cm), root length (10.96 cm), vigour index (2390), and germination percent (100 percent) in rice variety ADT-50.**

***Keywords* – Nano Zn Oxide, Synthesis, Characterization, Seed Priming, Rice.**

I. Introduction

Rice is a dominant crop being cultivated in India. Zinc deficiency in rice crops is termed Khaira disease and is characterized by interveinal chlorosis on the base of the new leaves and results in the formation of long white/ yellow streaks. Zinc acts as a cofactor or activator of many enzymes involved in carbohydrate metabolism, protein synthesis, maintenance of the integrity of the cellular membrane, regulation of auxin synthesis, and pollen formation. Zinc is reputed to be one of the important nutrients required for crop growth and animal health. The issue of micronutrient deficiency is related to food security (Meenakshi *et al*., 2010). Micronutrient deficiencies in human beings, as well as crop plants, are difficult to diagnose and consequently, the problem is termed 'hidden hunger' (Stein *et al*., 2007). Growing crops in Zn-deficient soil produces lesser yield with a poor zinc concentration of grain. The consumption of poor quality grain, low in zinc content develops zinc deficiency in the human population. In India, deficiency due to Zn is a major public health problem where people use more cereal-based food. Nanotechnology could be exploited to alleviate the Zn deficiency of humans through the biofortification of crops. The nanoparticles have a high surface area (30-50 m^2/g), high activity, better catalytic surface, rapid chemical reactions, and rapidly dispersible and absorb abundant water. Thus the use of nutrient particles in the nanometer range can serve as the potential alternative to overcome the limitation of presently available fertilizer. In the agronomic biofortification technique, priming of seeds is one of the important strategies which can efficiently increase the growth, yield, and nutrient content of crops. Earlier Slaton *et al.(2001*) demonstrated enhanced germination, root and shoot growth through priming of zinc nanoparticles.

II. Materials and Methods

ZnO nanoparticles were synthesized by direct precipitation method using 1M zinc acetate and 2M sodium hydroxide as precursors. The white precipitate was centrifuged and calcined at 500 °C as per the procedure of Ghorbani*et al*. (2015). The synthesized nanoparticles were characterized using the techniques of Fourier Transf-

-orm Infrared Spectroscopy (FTIR) as outlined by earlier workers (Geetha *et al*., 2016).

The seed priming experiment was carried out using four different rice varieties including two long-duration ADT 50 and CR 1009 and two short-duration ADT 45 and ADT 43 varieties procured from Tamil Nadu Rice Research Institute, Aduthurai. Seed priming on rice was done by soaking different varieties of rice seeds in deionized water for 6 hours followed by soaking them in the synthesized nano zinc solutions of varying concentrations viz., 200, 400, 600, 800, 1000, 1500, 2000 ppm for 6 hours and later rinsed thoroughly with water. After seed priming, the seeds were placed in between germination sheets and rolled. A control treatment without zinc priming was also studied. The measurement of root length, shoot length, germination percentage, and seed vigor index was undertaken. The best-performing rice variety and the standard zinc dosage were determined based on these measurements. The shoot and root length of all the germinated seeds were recorded in centimeters with the help of a measuring scale and the average shoot and root length were calculated. Germination percentage was calculated by taking the ratio of the number of seeds sown to the number of seeds germinated in a given time and expressed as a percentage. The seedling Vigour Index (SVI) was calculated by using the formula described by Abdul-Baki and Anderson (1973). Seed Vigour Index = Germination% × (Root length + Shoot length).

III. Results and Discussion

1. *Characterization of Zinc Oxide Nanoparticles -FT-IR Spectroscopy*

Figure 1 shows the FT-IR spectrum of the synthesized ZnO nanoparticle and it ranged from 400 to 4000 nm. The synthesized ZnO nanoparticle showed the absorption peaks at 430.79cm^{-1}, 480.87cm^{-1}, 558.821cm^{-1}, 594.74cm^{-1}, 712.62cm^{-1}, 899.81cm^{-1}, 1040.97, 1222.20cm^{-1}, 1368.08cm^{-1}, 1628, 1714cm^{-1}, 2853.67cm^{-1}, 2924.03cm^{-1} and 3444.81cm^{-1}. The bands of FT-IR spectroscopy peaks observed in the range of 2924 to 3444cm^{-1} correspond to O-H and C-H stretching. The band at 1368cm^{-1}corresponds to C-C stretching. The FT-IR band of ZnO nanoparticles at 430 to 594 cm^{-1} indicates Zn-O stretching. The presence of FT-IR peaks for ZnO nanoparticles and other metal oxides is well supported by previous literature (Hedayati, 2015; Gholamrezaei *et al.*, 2014).

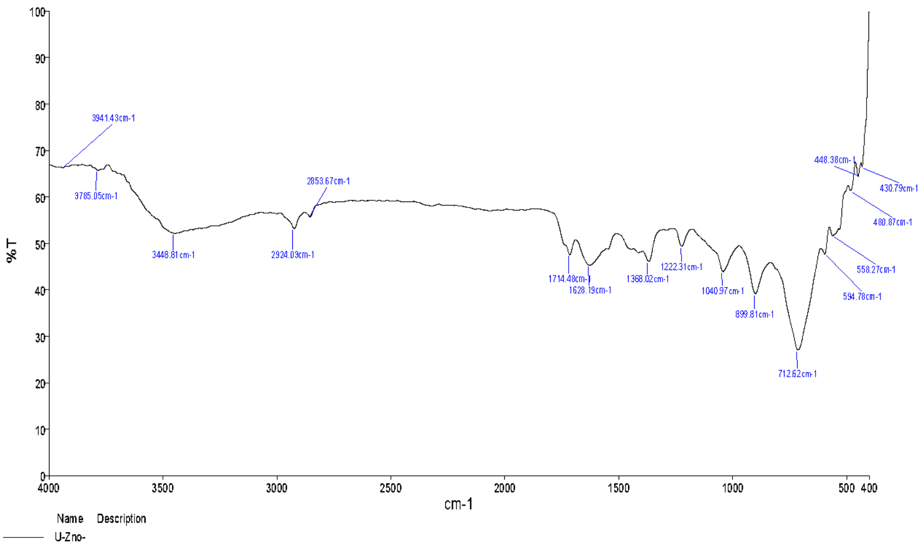

Fig. 1. The FT-IR spectroscopy pattern of ZnO nanoparticle.

2. *Effect of Nano Zinc Seed Priming on Rice*

2.1. *Shoot Length*

The results of the seed priming experiment indicated the significant influence of ZnO nanoparticles on the shoot length and root length of rice (Table 1). The shoot length of rice increased with increasing concentrations of zinc oxide seed priming up to 1000 ppm and then decreased. Among the varying concentration of zinc oxide seed priming, the treatment T_7 (1000 ppm) recorded the highest shoot length in all rice varieties evaluated. The 1000 ppm seed priming recorded a shoot length of 12.94 cm, 11.69 cm, 9.39 cm, and 7.69 cm on the 10th day by ADT- 50, ADT- 45, ADT- 43, and CR-1009 rice varieties respectively. The control treatment recorded the lowest shoot length in all the varieties. Among the varieties evaluated, ADT 50 was associated with the highest shoot length than other varieties. In ADT 50, the shoot length recorded in control on the 10th day was 7.14 cm. The shoot length increased to a range of 10.12-12.94 cm with zinc nano nutrient seed priming. Zinc plays an important role in the growth, nutrient composition, and oxidative enzyme activities of the plant (Aydin *et al.*, 2006). Zn as a metal constituent in enzymes or as a functional cofactor of a number of enzyme reactions played a role in the synthesis of tryptophan which is a precursor of the synthesis of IAA (Ali *et al.*, 2012). Increased shoot growth with nano Zn could also be attributed to the fact that there is an increase in the production of auxin hormone which enhanced the shoot growth. Zinc is used for protein synthesis, membrane function, cell elongation, and tolerance to environmental stresses (Cakmak, 2000). Earlier, Santo Pereira *et al.* (2021) showed that ZnO nanoparticles seed priming at low concentrations in enhancing the antioxidant enzymic and non-enzymic molecules, and phytohormones productivity thereby improved the initial growth of seedlings.

Table 1. Effect of Zno Nano nutrients on the shoot and root length of rice.

Treatments	**Shoot Length (cm)**				**Root Length (cm)**			
	ADT 50	**ADT 45**	**ADT 43**	**CR 1009**	**ADT 50**	**ADT 45**	**ADT 43**	**CR 1009**
T_1- Control	7.14	7.35	5.15	6.08	6.68	6.22	6.04	5.96
T_2-No soaking	8.52	8.02	5.97	6.51	7.19	7.02	6.53	6.21
T_3-200 ppm	10.78	8.83	6.77	6.97	7.97	7.21	6.62	6.59
T_4-400 ppm	10.12	9.01	8.17	7.02	8.16	8.38	6.84	6.81
T_5- 600 ppm	10.94	9.79	8.31	7.11	9.64	8.92	6.98	6.97
T_6-800 ppm	12.17	11.53	8.85	7.18	10.28	9.13	7.09	7.02
T_7- 1000 pm	12.94	11.69	9.39	7.69	10.96	9.82	7.89	7.64
T_8-1500 ppm	11.7	10.29	7.43	7.06	9.63	9.26	7.38	7.13
T_9- 2000 ppm	11.22	8.94	6.73	6.01	8.28	7.01	7.22	7.07
SEd	0.35	0.30	0.23	0.22	0.28	0.26	0.23	0.22
CD (p=0.05)	0.75	0.64	0.50	0.43	0.60	0.56	0.48	0.47

2.2. *Root Length*

The profound influence of zinc nano nutrient seed priming on increasing the root length of rice was well pronounced (Table1). Among the varieties of rice evaluated, ADT-50 (Long duration) and ADT-45 (short duration) ranked best by recording higher root length. Treatment T_7 ranked best in increasing the root length by recording 10.96, 9.82, 7.89, and 7.64 by ADT 50, ADT- 45, ADT-43, and CR-1009 varieties respectively. The concentration of Zn priming up to 1000 ppm was more useful than other concentrations evaluated. The control recorded the lowest root length of 6.68, 6.22, 6.04, and 5.96 cm by ADT-50 ADT- 45, ADT-43, and CR-1009 varieties respectively on the 10th day of treatment. Rice seeds showed differential growth in response to different concentrations of ZnO NPs. At concentrations higher than 1000 ppm, a reduction in shoot and root length was observed. The effect of nanoparticles at optimum dose is reported positive on the growth of plants. Therefore the concentration of ZnO NPs plays an important role in determining growth. Similar results were reported by Ramesh raddy *et al. (* 2017), where it was observed that seed treatment with ZnO at 1000 ppm showed an increase in root length and shoot length compared to the control. Results of Upadhyaya *et al.*(2017) revealed that Zinc nanoparticles (Zn NP) caused an increase in radicle and plumule length in rice. High seed Zn content in wheat has significantly improved the root and shoots growth under Zn deficient soils (Rengel and Graham 1995).

2.3. *Germination Percentage and Seed Vigour Index*

The influence of nano ZnO seed priming was significant in increasing the germination percentage of rice. While control recorded an average germination of 70 to 80 percent in different varieties, the various concentrations of ZnO nano treatments resulted in 90 to 100 percent germination. The seed priming with nano zinc caused 100 percent germination in all the seed varieties compared to the control treatment. This result was confirmed with Boonyanitipong (2011).

The Seed Vigour Index (SVI) of rice increased with increasing concentrations of zinc oxide seed priming upto 1000 ppm and then decreased at higher concentrations (Table 2). The treatment T_7 (1000 ppm) recorded the highest SVI in all rice varieties evaluated. The 1000 ppm seed priming recorded an SVI of 2390, 2151, 1728, and 1533 on the 10th day by ADT-50, ADT-45, ADT-43, and CR-1009 rice varieties respectively. The control treatment recorded the lowest SVI of 1243.8, 1246.5, 1023.3, and 1090.8 on the 10th day by ADT-50, ADT-45, ADT-43, and CR-1009 rice varieties respectively. The other concentrations of nano Zn also significantly increased the seed vigour index compared to the control.

Table 2. Effect of Nano Zinc on the germination percentage and vigour index of rice.

Treatments	**Germination percentage**				**Vigour Index**			
	ADT 50	**ADT 45**	**ADT 43**	**CR 1009**	**ADT 50**	**ADT 45**	**ADT 43**	**CR 1009**
T_1- Control	80	90	90	90	1105.6	1221.3	1007.1	1083.6
T_2-No soaking	90	90	90	90	1413.9	1353.6	1125.0	1144.8
T_3-200 ppm	90	100	100	90	1687.5	1604.0	1339.0	1220.4
T_4-400 ppm	100	100	100	100	1828.0	1739.0	1501.0	1378.0
T_5- 600 ppm	100	100	100	100	2058.0	1871.0	1529.0	1408.0
T_6-800 ppm	100	100	100	100	2245.0	2066.0	1594.0	1420.0
T_7- 1000 pm	100	100	100	100	2390.0	2151.0	1728.0	1533.0
T_8-1500 ppm	100	100	100	100	2133.0	1955.0	1481.0	1419.0

Treatments	Germination percentage				Vigour Index			
	ADT 50	ADT 45	ADT 43	CR 1009	ADT 50	ADT 45	ADT 43	CR 1009
T_9- 2000 ppm	1000	100	90	90	1950.0	1595.0	1255.5	1177.2
SEd	3.15	3.23	3.21	3.10	63.24	58.49	45.95	42.70
CD (p = 0.05)	6.68	6.85	6.54	6.57	132.87	122.88	97.41	90.52

The possible reasons for the control treatment to be significantly lower compared to seed priming treatments may be due to the fact that zinc may affect plant water relations and alter stomatal conductance. Stomatal conductance and transpiration rates also declined under zinc deficiency. Gas exchange data presented by Sharma *et al*. (1994) indicated that zinc deficiency causes a reduction in the instantaneous transpiration efficiency of leaves. The seedlings emerging from low Zn seeds showed poor seedling vigor and signifying the importance of maintaining optimum Zn levels (Yilmaz et al. 1998). Seed priming with Zn in barley seeds was effective in improving seed germination and seedling development (Ajouri et al. 2004). The fact that priming seed with Zn was effective in improving seedling growth, indicates that the prime Zn that accumulated in the husk was much more accessible to meet the immediate need of the germinating seed than soil-applied Zn (Prom-U-Thai *et al.*, 2012). The present finding also corroborates the report of Prabha Raj-Kalal and Anjana Jajoo (2021), they demonstrated that wheat seeds primed with ZnO NPs (10 mg/L) showed a significant positive influence on seed germination performance and vigour index through enhanced seed water uptake resulting in an enhanced α-amylase activity.

IV. Conclusion

The results of the seed priming experiment indicated the significant influence of Zn nanoparticles on increasing the root length and shoot length of rice. The root and shoot length of rice in all the varieties increased with increasing concentrations of zinc oxide seed priming up to 1000 ppm. Among the varieties evaluated, ADT 50 was associated with the highest shoot and root length than other varieties. In ADT 50, the shoot length increased to a range of 10.78-12.94 cm and root length to a range of 7.97-10.96 cm with zinc nano nutrient seed priming as with nano ZnO seed priming. The treatment T_7 (1000 ppm ZnO) recorded the highest Seed vigour index in all rice varieties evaluated.

References

[1] Abdul-Baki and J.D. Anderson, Vigour determination in Soya bean seed by multiple criteria, Crop Sci., 1973, 13(6). pp. 630-633.
[2] R.I. Ali, T.H. Awan, M. Ahmed, M.U. Saleem and M. Akhtar, Diversification of rice based cropping system to improve soil fertility, sustainable productivity and economics, J. Animal and Plant Sciences, 2012, **22**.pp.105-112.
[3] A. Ajouri, H. Asgedm and M. Becker, Seed priming enhances germination and seedling growth of barley under conditions of P and Zn deficiency. Journal of Plant Nutrition and Soil Sci, 2004, 167(5). pp. 630-636.
[4] M. Aydin, M.A. Demiral and M.A. Kaptan, Effect of zinc on growth, nutrient composition and anti-oxidative enzyme activity of maize as influenced by phosphorus, Int. J. Biol. Biotech., 2012, 3. pp. 411-417.
[5] P. Boonyanitipong, B. Kositsup Kumar, S. Bauah and J. Dutta, Toxicity of ZnO and TiO_2 nanoparticles on germinating rice seed Oryza Sativa. Int J. Biosci Biochem Bioinform,2011,1, pp.*285- 85.*
[6] I. Cakmak, Role of Zinc in protecting plant cells from reactive oxygen species, Newphytol, 2000,146,pp. 185-205.
[7] M. Faizan, A. Faraz, M. Yosuf , S.T. Khan, and S. Hayat, Zinc oxide nanoparticle mediated changes in photosynthetic efficiency and antioxidant system of tomato plants. Photosynthetica, 2018,56(2).pp.678-686.
[8] M.S. Geetha, H. Nagabhushana, and H.N. Shivananjaiah, Green mediated synthesis and characterization of ZnO nanoparticles using Euphorbia Jatropha latex as reducing agent. Journal of Science: Advanced Materials and Devices, 2016,1,pp. 301-310.
[9] S. Gholamrezaei, M. Salavati-Niasari, and D. Ghanbari. J Indus Eng Chem, 2014, 20.pp.3335-3341.
[10] H.R. Ghorbani, F.P. Mehr, H. Pazzoki, and B.M., Rahmani, Synthesis of ZnO nanoparticles by precipitation method, Oriental Journal of Chemistry. 2015. 31(2), pp.1219-1221.
[11] K. Hedayati, Fabrication and Optical Characterization of Zinc Oxide, Nanoparticles Prepared via a Simple Sol-gel Method, Journal of nanostructures,2015,5, pp. 395-401.
[12] J.V. Meenakshi, Nancy Johnson, V.M. Manyong, and Hugo De Groote, How cost-effective is bio fortification in combating malnutriti-

-on? An Ex Ante assessment. World Devl., 2010.38(1),pp. 67-75.

[13] C. Prom-u-thai, Benjavan Rerkasem, Atilla Yazici, and Ismail Cakmak, Zinc priming promotes seed germination and seedling vigor of rice, *J. Plant Nutr. Soil Sci.*2012.*175, pp.*482–488.

[14] Prabha Rai-Kalal and Anjana Jajoo, Priming with zinc oxide nanoparticles improve germination and photosynthetic performance in wheat, Plant physiology and biochemistry ,2021,160.pp.341-351.

[15] Ramesh Reddy, G.J. Pavithra, B.H. Rajashekar Reddy, Mahesh Salimath, K.N. Geetha and A.G. Shankar.2017. Zinc oxide nanoparticles increase Zn uptake, and translocation in rice with a positive effect on growth, yield, and moisture stress tolerance. Ind.J. Plant Physiol. DOI 10.1007/s40502-017-0303-2.

[16] Z. Rengel and R.D.Graham. Importance of seed zinc content for wheat growth on zinc deficient soils, I. Vegetative growth. *Plant and soil*. 1995, 173,pp. 259- 266.

[17] A. Santo Pereira, .D.E.; Oliveira, H.C.; Fraceto, L.F.; Santaella, C. Nanotechnology Potential in Seed Priming for Sustainable Agriculture. Nanomaterials, 2021, 11, 267. [Cross Ref] [Pub Med].

[18] P.N. Sharma, N. Kumar, SS. Bisht. Effect of zinc deficiency on chlorophyll contents, photosynthesis and water relations of cauliflower plants. Photosynthetic.1994, 30, pp.353.

[19] N.A. Slaton, N.A, C.E. Wilson Jr., S. Ntamatugiro, R. J. Norman, and D.L. Boothe, Evaluation of zinc seed treatment for rice, Argon J., 2001, 93, pp.157- 163.

[20] A.J. Stein,A.J ., P. Nestel, J.V. Meenakshi, M. Quain, H.P.S. Sachdev and Z.A. Bhutta, Plant breeding to control zinc deficiency in India: How cost-effective is biofortification? Public Health Nutrition, 2007. 10(5), pp. 492-501.

[21] H.H. Upadhyaya, S. Roy, S. Shome, Tewari, and M.K. Bhattacharya. The physiological impact of Zinc nanoparticle on germination of rice (*Oryza sativa L*) seed, J. Plant SciPhytopathology, 2017, 1, pp. 062-070.

[22] A. Yilmaz, H. Ekiz, I. I. Gultekin, B. Torun, H. Barut, S. Karanlik and I. Cakmak, Effect of seed zinc content on grain yield and zinc concentration on wheat growth on zinc-deficient calcareous soil, J. Plant nutrition. 1998, 21, pp. 2257-2264.

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