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Effect of seed priming with biosynthesized iron nanoparticles on seed germination and seedling characteristics of wheat

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Abstract
The lab experiment was conducted to standardize the concentration of biosynthesized iron nanoparticles (Fe NPs) for seed priming in wheat, which was synthesized through Actinobacteria and *Pseudomonas* were carried out at Green nanotechnology lab, UAS, Dharwad, Karnataka during 2021-22 in completely randomized design with three replications. The results revealed that, wheat seeds primed with biosynthesized Fe NPs through Actinobacteria (T_6) and *Pseudomonas* (T_1) at 250 ppm recorded significantly higher germination (97.00 and 96.67%), root length (15.62 and 14.80 cm), shoot length (12.83 and 12.57 cm) and seedling vigour index (2759.65 and 2645.86) respectively, which was on par with Fe NPs seeds priming at 500 ppm biosynthesized through Actinobacteria (T_7) and *Pseudomonas* (T_2) when compared to control (87.00%, 11.87 cm, 9.80 cm and 1885.29, respectively) (T_{11}).

Keywords: Biosynthesis, iron, nanoparticles, seed priming, wheat

Introduction
Wheat (*Triticum aestivum* L.), a major staple food crop cultivated globally, is a self-pollinating long-day plant of the Poaceae family, providing around 35% of the total food consumed by the world's population. The majority of wheat that is grown on a worldwide is hexaploid, and it is widely utilised to make a variety of baked food products including bread (Mohammadi-joo *et al.*, 2015) [8].

Nanotechnology may help bring about a new technological revolution in agriculture. It is possible to produce nanofertilizers using nanomaterials because of their high surface-to-volume ratio, gradual and controlled release at target places, and other characteristics (Shakiba *et al.* 2020) [10]. Additionally, nanofertilizers, as opposed to chemical fertilisers, allow for high mineral bioavailability to plants due to their smaller size, greater reactivity, and higher surface area (Liu and Lal, 2015) [7].

Synthesis of nanoparticles involves a variety of biological, physical and chemical methods; however, only the biological methods are safe and environment friendly. Traditional chemical methods, which could be hazardous to the environment, are being replaced with biological systems, which are preferred for nanoparticles synthesis (Busi and Rajkumari, 2019) [5]. Microorganisms (fungi, viruses, bacteria, yeast, and actinomycetes) are great producers of nanoparticles because they can create a variety of secondary metabolites (Ovais *et al.*, 2018) [9].

Iron (Fe), an essential nutrient for the crop growth and development of crops. It is essential for the formation of chlorophyll and is involved in the electron transport system and activation of several enzymatic functions (Ali *et al.*, 2021) [3]. In order to increase productivity and the quality of the food produce, seed priming has been used to synchronise and speed up germination, boost seedling vigour, and increase plant resistance to biotic and abiotic stresses (Carrillo-Reche *et al.*, 2018) [6].

Materials and Methods
Purpose of lab experiment is to standardize the concentration of biosynthesized iron nanoparticles. Collection and screening of *Pseudomonas* and Actinobacterial isolates were done at Microbial Genetics Laboratory, Department of Agricultural Microbiology, UAS, Dharwad.

Biosynthesis, characterization and standardization of Fe nanoparticles, the lab experiment were done in Green Nanotechnology Laboratory, University of Agricultural Sciences, Dharwad, Karnataka during 2021-22. This experiment was laid out in completely randomized design (CRD) with 11 treatments and three replications. Wheat seeds of variety UAS 334 were soaked in biosynthesized iron nanoparticles (Fe NPs) solution at 250, 500, 750, 1000 and 1500 ppm concentration for a period of 6 hours. T_1-Seed priming with BS Fe NPs @ 250 ppm, T_2-Seed priming with BS Fe NPs @ 500 ppm, T_3- Seed priming with BS Fe NPs @ 750 ppm, T_4-Seed priming with BS Fe NPs @ 1000 ppm, T_5-Seed priming with BS Fe NPs @ 1500 ppm, T_6- Seed priming with ABS Fe NPs @ 250 ppm, T_7-Seed priming with ABS Fe NPs @ 500 ppm, T_8-Seed priming with ABS Fe NPs @ 750 ppm, T_9-Seed priming with ABS Fe NPs @ 1000 ppm,T_{10}-Seed priming with ABS Fe NPs @ 1500 ppm, T_{11}-Hydropriming.

Observations recorded

Seed germination

Germination test was conducted using three replicates of 100 seeds each in rolled paper towel and incubated in the walk-in seed germination room at 25 ± 2 °C temperature and 90 ± 5 percent RH. Seedling evaluation was done when seedlings have reached a stage with all the essential structures were fully expressed. Sufficient time was given for the seeds to germinate and produce all essential structures showing potentiality to develop into normal plant under favourable conditions. Such seedlings were considered as normal seedlings and counted to compute the germination percentage. The number of normal seedlings in each replication was counted at the end of 8th day and the germination percentage was calculated and was expressed in percentage as given by ISTA (Anon, 2011) [4].

$$\text{Normal seedlings (\%)} = \frac{\text{Number of normal seedlings}}{\text{Total number of seeds}} \text{ x } 100$$

Root length

From the germination test, ten normal seedlings were selected randomly in each treatment from each replication on 8th day. The root length was measured from the tip of the primary root to hypocotyl and mean root length was expressed in centimetre.

Shoot length

From the germination test, ten normal seedlings were selected randomly in each treatment from each replication on 8th day. The shoot length was measured from the base of the primary leaf to hypocotyl and mean shoot length was expressed in centimetre.

Seedling vigour index (SVI)

Vigour index was computed by using the following formula and expressed in number (Abdul-Baki and Anderson, 1973) [1].

Seedling vigour index = Germination % × [Shoot length (cm) + Root length (cm)].

Results and Discussion

The data on germination as influenced by seed priming with different concentrations (250, 500, 750, 1000 and 1500 ppm) of biosynthesized Fe NPs using microbial extract of *Pseudomonas* and Actinobacteria is given in the Table 1.

Wheat seeds primed with biosynthesized Fe NPs through Actinobacteria (T_6) and *Pseudomonas* (T_1) at 250 ppm recorded significantly higher germination percent (97.00 and 96.67%), root length (15.62 and 14.80 cm), shoot length (12.83 and 12.57 cm) and seedling vigour index (2759.65 and 2645.86) respectively, which was on par with Fe NPs seeds priming at 500 ppm biosynthesized through Actinobacteria (96.67%, 14.83 cm, 12.23 cm and 2615.89, respectively) (T_7) and *Pseudomonas* (95.33%, 14.40 cm, 11.92 cm and 2509.09, respectively) (T_2) (Fig 1). Significantly lower seed germination, root length, shoot length, and seedling vigour index (9.80 cm) (T_{11}) was recorded in control (87.00%, 11.87 cm, 9.80 cm and 1885.29, respectively) (T_{11}). Seed priming with iron nanoparticles significantly enhanced seed germination and seedling growth in wheat, and that the α-amylase activity of nano treated seeds increased in comparison to unprimed seeds. Increased activity of α-amylase allowing for a higher rate of starch hydrolysis in germinating nano-primed seeds might be the cause of an increase in germination rate and seedling vigour in nano-primed seeds by Yasmeen *et al.* (2015) [11]. Iron nanoparticles priming has a considerable impact on wheat seed germination, root length, and shoot length. Iron nanoparticles alter physiological and biochemical processes, which affects their germination and development (Alam *et al.*, 2015) [2].

Table 1: Effect of seed priming with biosynthesized Fe NPs on seedling characteristics of Wheat

Treatments	Germination (%)	Root length (cm)	Shoot length (cm)	Seedling vigour index
T_1-SP with BS Fe NPs @ 250 ppm	96.67^a	14.80^a	12.57^a	2645.86^a
T_2-SP with BS Fe NPs @ 500 ppm	95.33^a	14.4^a	11.92^a	2509.09^a
T_3-SP with BS Fe NPs @ 750 ppm	89.67^{bc}	12.53^b	10.43^b	2058.82^b
T_4-SP with BS Fe NPs @ 1000 ppm	89.00^{bc}	12.25^b	10.25^b	2002.50^b
T_5-SP with BS Fe NPs @ 1500 ppm	87.33^c	11.93^b	9.85^b	1902.05^b
T_6-SP with ABS Fe NPs @ 250 ppm	97.00^a	15.62^a	12.83^a	2759.65^a
T_7-SP with ABS Fe NPs @ 500 ppm	96.67^a	14.83^a	12.23^a	2615.89^a
T_8-SP with ABS Fe NPs @ 750 ppm	90.00^{bc}	13.10^b	10.67^b	2139.30^b
T_9-SP with ABS Fe NPs @ 1000 ppm	89.33^{bc}	12.37^b	10.35^b	2029.58^b
T_{10}-SP with ABS Fe NPs @ 1500 ppm	87.67^{bc}	12.13^b	10.13^b	1951.53^b
T_{11}-Hydropriming	87.00^c	11.87^b	9.80^b	1885.29^b
S.Em.±	0.78	0.31	0.23	65.34

SP-Seed priming; BS-Bacterial (*Pseudomonas*) synthesized; ABS-Actinobacterial synthesized

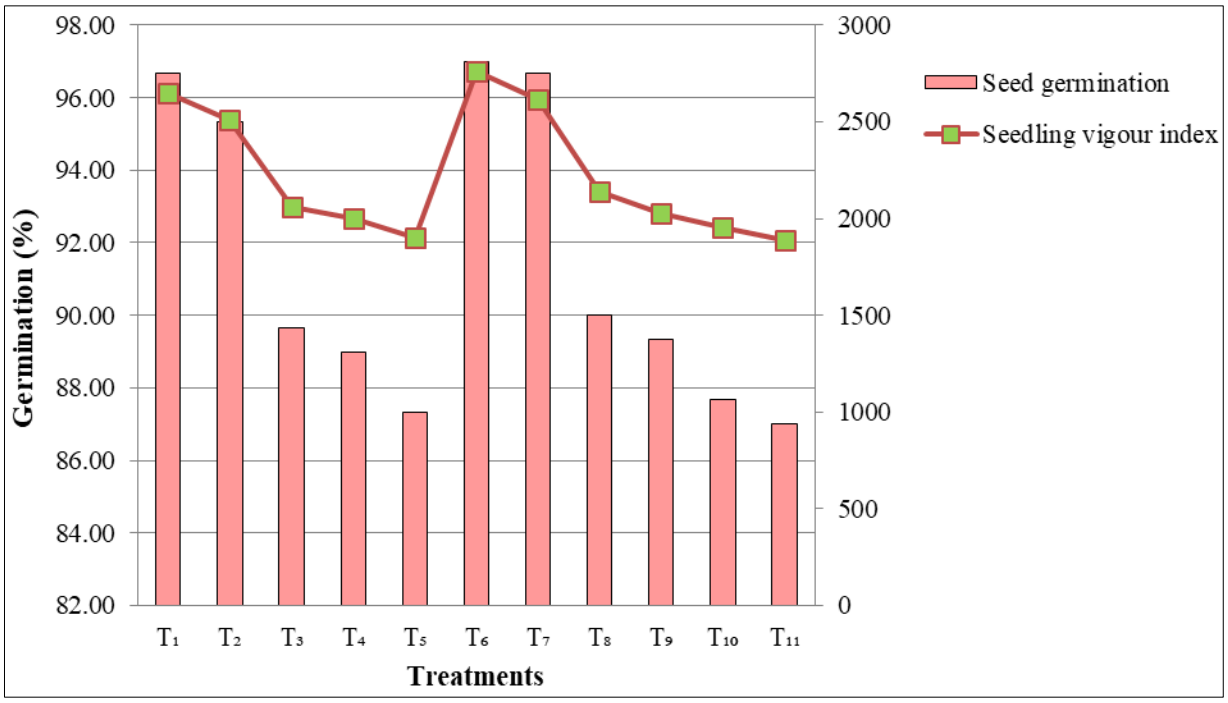

Fig 1: Effect of seed priming with biosynthesized Fe NPs on seed germination and seedling vigour index in wheat

Conclusion

Wheat seeds primed with 250 ppm of iron nanoparticles biosynthesized using Actinobacteria and *Pseudomonas* shown superior in seed germination, root length, shoot length and seedling vigour index. Seed priming has been effectively influenced the vigour and stand establishment of seedling with biosynthesized iron nanoparticles.

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