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EXPLORATION OF ANTIFUNGAL PROPERTY OF MANGANESE NANOPARTICLES AGAINST *Sclerotium rolfsii* A PHYTOPATHOGEN OF SOYBEAN (*Glycine max*, L. Merrill)

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ABSTRACT
Soybean (*Glycine max* (L.) Merril) is a one of the most important oil crop in Maharashtra. Fungal phytopathogens are most important among the biotic factors that cause serious losses to agricultural crops. *Sclerotium rolfsii* is a plant pathogenic fungus frequently infected Soybean crop and leads into decrease crop productivity. The present research is concerned with the antifungal property of manganese nanoparticles used against the treatment of Soybean plant pathogenic fungi. We used manganese nanoparticles at concentrations of 00, 25, 50, 75 and 100 ppm. *Sclerotium rolfsii* a Soybean plant pathogenic fungus was used as test pathogens and treated with manganese nanoparticles. The experiment was carried out by adopting sick pot technique. We calculated Germination %, Root-Shoot Length, Seedling Vigor Index and Disease Incidences in order to evaluate the antifungal efficacy of manganese nanoparticles against fungal pathogens. The results indicated that manganese nanoparticles possess antifungal properties against these plant pathogens at various levels. Treatment with 50 ppm concentration of manganese nanoparticles resulted in maximum inhibition of test fungi. Results also showed that the inhibition property was decreased with the concentration of 25, 75 and 100 respectively. In summary, manganese nanoparticles showed potent antifungal effects on *Sclerotium rolfsii*. Thus, it can be effectively used against plant phytopathogenic fungi to protect the Soybean crop and their products, instead of using the commercially available synthetic fungicides, which show higher toxicity to humans. These results suggest that manganese nanoparticles could be used as an effective fungicide in agricultural and food safety applications.

KEYWORDS: Manganese nanoparticles, Antifungal property, Soybean crop and *Sclerotium rolfsii*.

INTRODUCTION
Nanotechnology is the manipulation or self-assembly of individual atoms, molecules, or molecular clusters into structures to create materials and devices with new or vastly different properties. The potential of nanotechnology to revolutionize the health care, textile, materials, information and communication technology, and energy sectors has been well-publicized. The magnetic properties of manganese oxide nanoparticles are of increasing research interest due to their intrinsic high atomic moment.[1] Manganese oxides can be applied in catalysts, molecular-sieves, ion-sieves, batteries, magnetic materials as well as other applications such as water treatment, imaging contrast agents due to their excellent physicochemical properties[2, 3] observed inhibition activity MnNPs against two gram-positive bacteria viz. *Staphylococcus aureus* and *Bacillus subtilis*, two gram negative bacteria Viz. *Escherichia coli* and *Staphylococcus bacillus* and antifungal activity against four funguses Viz *Candida albicans*, *Curvularia lunata*, *Aspergillus niger* and *Trichophyton simii*. Recent studies have demonstrated antimicrobial activities of various nanoparticles materials, including silver[4-5], copper[6], titanium dioxide[7], and zinc oxide.[8] The uses of nanoparticles of have been considered alternate, ecofriendly and cost effective management strategy for the control of pathogenic microbes.[9-10] These nanoparticles have a great potential in the management of plant diseases as compared to synthetic fungicides.[11] Thus, use of nanoparticles has been considered an alternate and effective approach which is eco-friendly and cost effective for the control of pathogenic microbes.[10] These nanoparticles have a great potential in the management of plant diseases as compared to synthetic fungicides.[11] Antimycotic activity of some nanoparticles has also been reported on some fungi like wood rotting fungi, *Fusarium* species and other phytopathogenic fungi.[12] Biological control methods are perceived to have specific advantages over synthetic fungicides because of less non target and environmental

effects. They also shows efficacy against fungicide-resistant pathogens and reduced probability of resistance development. However, the less information is available on antimycotic activity of Manganese nanoparticles. Hence, in the present study, the antifungal property of Manganese nanoparticles against *Sclerotium rolfsii* a phytopathogen of Soybean plant will be investigated.

MATERIALS AND METHODS

The studies on effect of Manganese nanoparticles on diseases incidences of Soybean plants were carried out by adopting sick pot technique.[13] *S. rolfsii* isolated from diseased Soybean plant was used for preparation of fungal pathogen sick soil as described by[13] with slight modification. Manganese nanoparticles were synthesized in previous studies by using fungi and characterized by standard methods. Soybean seeds were coated with Manganese nanoparticles at various concentrations. Manganese nanoparticles were diluted to different concentration viz., 25 ppm, 50 ppm, 75 ppm and 100 ppm in deionized distilled water and sonicated for 30 min.[14] The seeds were surface sterilized and were soaked separately with Manganese nanoparticle solution for 45 min and removed and air dried at room temperature in laboratory. Seed coated without nanoparticles were maintained as control. 25 treated seed of soybean cultivar JSB-335 were dibbled manually in sick soil pots. Pots with untreated seeds were kept as control. All the pots were irrigated and kept in nursery upto 30 DAS. Germination count was recorded on 10^{th}DAS. Three plants from each plot were selected randomly and tagged for recording different biometric observation *viz,* Germination percent, root-shoot length, and Seedling Vigor Index (SVI) at 30^{th} DAS. Mean of three plants were considered for analysis.

Seedling Vigour Index (SVI) was calculated by the formula.[15]

SVI = (root length + shoot length) × Germination per cent

Number of healthy and infected plant were evaluated and percent disease incidences was studied at 30^{th} days of sowing and calculated as suggested by.[16]

$$Disease\ Incidences = \frac{\text{No. of infected plant units}}{\text{Total No. of (Healthy + Infected) plant unit}}$$

RESULT AND DISCUSSION

Germination percentage

Results on the influence of manganese nanoparticle treatments on germination percent are presented in Table (1) and graphically represented in Fig (1). From the table it is evident that as the level of Manganese nanoparticle increases the germination percentage was also increased but up to 50 ppm level of Manganese nanoparticle and decreased thereafter with further increase in the concentration of nanoparticle. Maximum germination percent (72.00) per cent was recorded at 50 ppm followed by 25 ppm at which the germination recorded was (61.32) per cent which was significantly superior over control at which the germination was only (9.32) percent. It was also observed that germination percentage at 75 ppm (50.64) and 100 ppm (32.00) was numerically different. However both the treatments were statistically at par which each other for seed germination. The results revealed that treatments with manganese nanoparticle significantly improved the germination of soybean over untreated control.

Root Length

Results on the influence of manganese nanoparticle treatments on root length of soybean in *S. rolfsii* sick soil are presented in Table (1) and graphically represented in Fig (2). From the result it was observed that the treatment of manganese nanoparticles increases the length of root. The numerical increase in root length was found to be increased with increased in concentration of manganese nanoparticles up to 50 ppm and decreased thereafter. The maximum root length (6.92 cm) was recorded at 50 ppm, followed by 25 (5.80 cm), 75 ppm (5.58 cm) and the minimum root length (4.56 cm) was recorded in treatment with 100 ppm Manganese nanoparticle and all the treatments were found to be statistically significant for root development over control at which it was only (4.11 cm).

Shoot Length

Results on the influence of manganese nanoparticle treatments on shoot length of soybean in *S. rolfsii* sick soil are presented in Table (1) and graphically represented in Fig (2). From the result it was observed that the treatment of manganese nanoparticles increases the length of shoot. The numerical increase in shoot length was found to be increased with increased in concentration of manganese nanoparticles up to 50 ppm and decreased thereafter. The maximum shoot length (19.95 cm) was recorded at 50 ppm, followed by 25 (16.19 cm), 75 ppm (15.50 cm) and the minimum shoot length (15.07 cm) was recorded in treatment with 100 ppm Manganese nanoparticle and all the treatments were found to be statistically significant for shoot development over control at which it was only (14.35 cm).

Seedling Vigor Index

Results on the influence of manganese nanoparticle treatments on Seedling Vigor Index of soybean in *S. rolfsii* sick soil are calculated and presented in Table (1) and graphically represented in Fig (3). From the result it was observed that the treatment of manganese nanoparticles shows increase in the Vigor of Seedling. The maximum Vigor (1934.64) was recorded at 50 ppm, followed by 25 (1348.42), 75 ppm (1067.49) and the minimum Seedling Vigor Index (628.16) was recorded in treatment with 100 ppm Manganese nanoparticle and all the treatments were found to be statistically significant for Seedling Vigor Index over control at which it was only (172.04).

Disease Incidences in Soybean

Results on the influence of manganese nanoparticle treatments on per cent disease incidences of soybean in

S. rolfsii sick soil are calculated and presented in Table (2) and graphically represented in Fig (4). From the result it was observed that, the minimum disease incidences (14.77) per cent was recorded at 50 ppm, followed by treatment 25 (41.29) per cent, in 75 ppm treatment it was (55.29) per cent and the maximum disease incidences (83.25) per cent was recorded in treatment with 100 ppm Manganese nanoparticle. The results revealed that, the treatments with manganese nanoparticle significantly reduce the disease incidences of soybean over untreated control (85.83), the study enlights the possible use of Manganese nanoparticle for plant disease control agent against *S. rolfsii*.

Similar reports were also recorded by[17], who found that significant effects of nanosized TiO2 on spinach seed germination. They referred results to small particle size, which permitted nanoparticles to penetrate into the seed during the treatment period, enhancing their functions throughout the growth period.

Table 1: Effect of graded doses of Manganese nanoparticles on germination and Seedling Vigor Index (SVI) in soybean at 30 DAS in *S. rolfsii* sick soil.

Treatment	Mean Germination count	Mean Root Length	Mean Shoot Length	Germination %	Seedling Vigor Index (SVI)
00 (Control)	2.33	4.11	14.35	9.32	172.04
25 ppm	15.33	5.80	16.19	61.32	1348.42
50 ppm	18.00	6.92	19.95	72.00	1934.64
75 ppm	12.66	5.58	15.50	50.64	1067.49
100 ppm	8.00	4.56	15.07	32.00	628.16
F-test	**Sig**	**Sig**	**Sig**		
SE.(m) ±	0.760117	0.117222	0.342812		
C.D.(P=0.05)	2.395028	0.369353	1.080155		

Table 2: Effect of graded doses of Manganese nanoparticles on disease incidences in soybean at 30 DAS in *S. rolfsii* sick soil.

Treatment	Mean Number of Infected Plant Units	Mean Number of Total Plant Units	%D.I.
00 (Control)	2.00	2.33	85.83
25 ppm	6.33	15.33	41.29
50 ppm	2.66	18.00	14.77
75 ppm	7.00	12.66	55.29
100 ppm	6.66	8.00	83.25
F-test	**Sig**	**Sig**	
SE.(m) ±	0.679052	0.760117	
C.D.(P=0.05)	2.139602	2.395028	

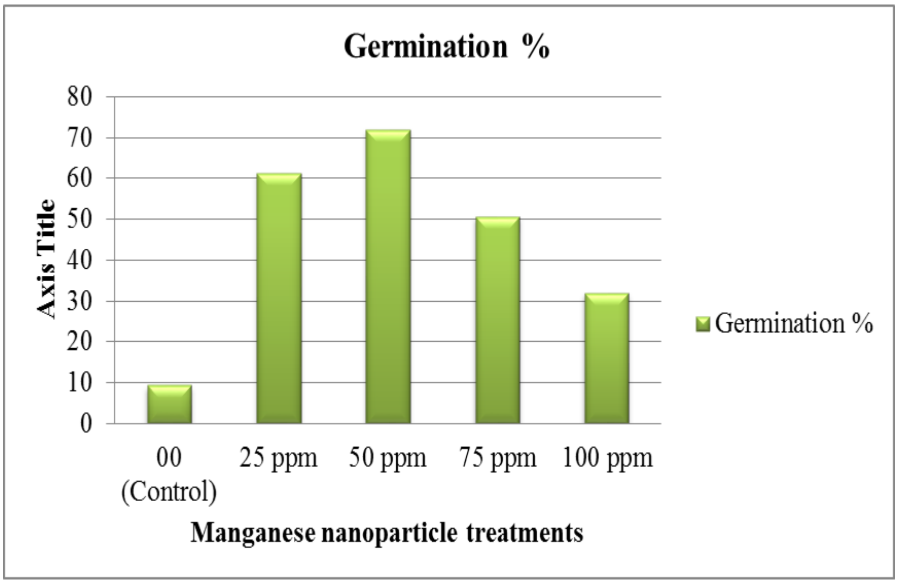

Figure 1: Effect of graded doses of Manganese nanoparticles on Germination of soybean at 30 DAS in *S. rolfsii* sick soil.

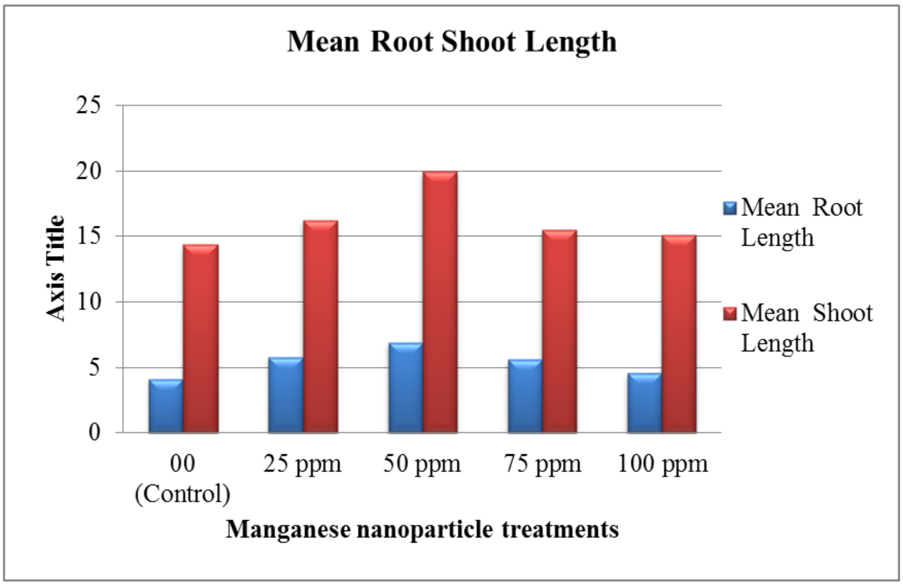

Figure 2: Effect of graded doses of Manganese nanoparticles on Root Shoot Length of soybean at 30 DAS in *S. rolfsii* sick soil.

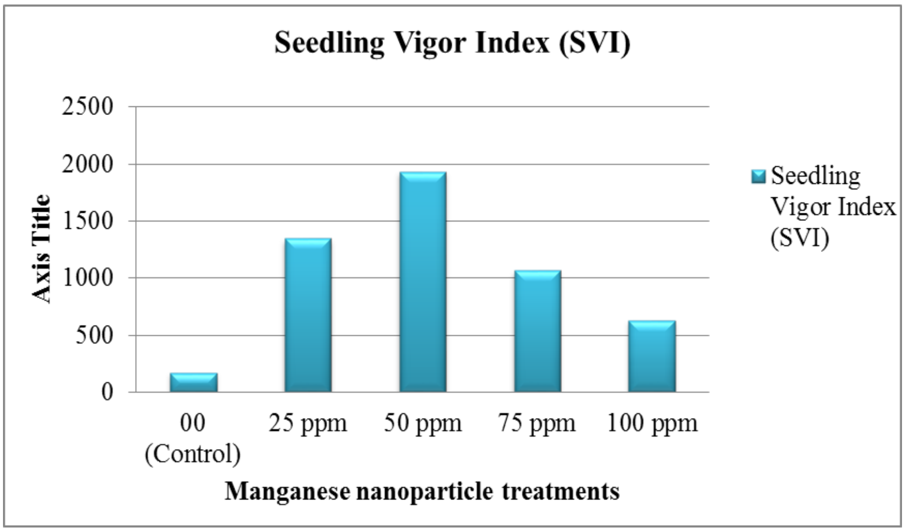

Figure 3: Effect of graded doses of Manganese nanoparticles on Seedling Vigor Index (SVI) of soybean at 30 DAS in *S. rolfsii* sick soil.

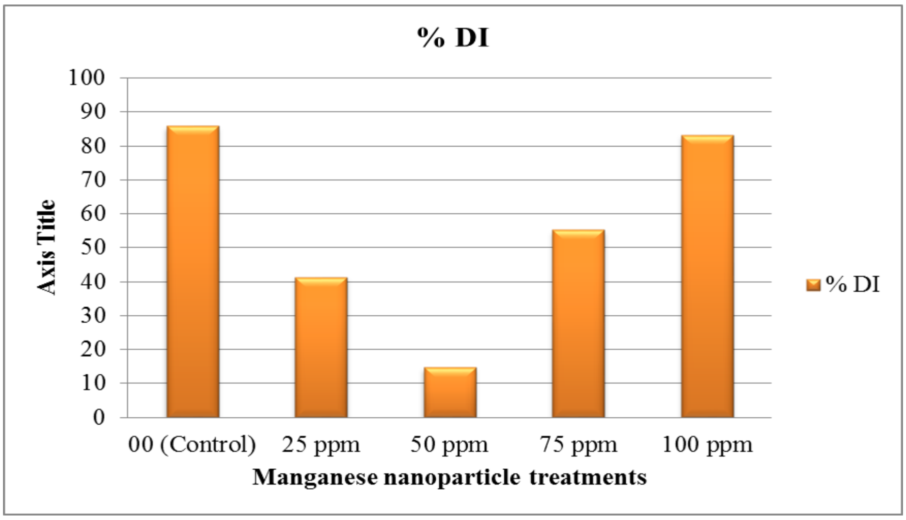

Figure 4: Effect of graded doses of Manganese nanoparticles on disease incidences of soybean at 30 DAS in *S. rolfsii* sick soil.

CONCLUSION

Management of fungal diseases of Soybean crops is economically important. Recently, a greater effort has been given to development of safe management methods that pose less danger to humans and animals, with a focus on overcoming deficiencies of synthetic fungicides. Findings from the current investigation demonstrated that manganese nanoparticles were very effective against plant phytopathogenic fungi. However, the present study is based on adopting sick pot technique. The finding provides valuable information on manganese nanoparticles for its use in control of plant pathogens. In this study, we analyzed the inhibition effect of manganese nanoparticles against Soybean plant pathogenic fungi *Sclerotium rolfsii*. The results suggest that manganese nanoparticles are capable of inhibiting these pathogens. *Sclerotium rolfsii* showed a high inhibition effect at a 50 ppm concentration of manganese nanoparticles. In summary, manganese nanoparticles exerted potent antifungal effects on test fungi, probably through destruction of membrane integrity; therefore, it was concluded that manganese nanoparticles have considerable antifungal activity. Thus, it can be effectively used against plant phytopathogenic fungi to protect the various crop plants and their products, instead of using the commercially available synthetic fungicides, which show higher toxicity to humans. Moreover, this report opens up for further research, the area of mode of action of manganese nanoparticles on the phytopathogenic fungi. Further investigation for mass applications is needed.

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