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The effect of beneficial microbial bioagents on plant growth metrics

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ABSTRACT

In the current study titled *" The effect of* beneficial microbial bioagents on plant growth metrics*,"* the bioagents *Trichoderma harzianum*, *Bacillus subtilis* and *Pseudomonas fluorescens* were tested along with seed dressing broad spectrum fungicide tebuconazole @1.0 % to know the effect of beneficial bioagents on plant growth metrics like germination per cent, vigor index, shoot and root length under *in vitro* conditions. Seed treatment with *T. harzianum* (T_1) significantly enhanced germination percentage (98.5%), shoot and root lengths (18.97 cm and 16.50 cm) and vigor index (3495.20), outperforming all other treatments. *B. subtilis* (T_3) and the chemical control (T_4) also showed positive effects, while the untreated control (T_5) consistently recorded the lowest values. These results highlight *T. harzianum* as the most effective treatment for promoting early plant growth. In case of seeds pretreated with the *S. rolfsii*, chemical control (T_4) showed the highest germination (86.0 %), shoot and root lengths (15.90 cm and 15.17 cm) and vigor index (2672.45), followed by *T. harzianum* (T_1). The pathogen control (T_5) consistently recorded the lowest values, indicating the effectiveness of tebuconazole and *Trichoderma* in promoting early plant growth. Similar results were observed in case of seeds pretreated with *M. phaseolina*, chemical control (T_4) showed the highest germination (87.0 %), shoot and root lengths (15.32 cm and 13.97 cm) and vigor index (2672.45), followed by *T. harzianum* (T_1). The pathogen control (T_5) consistently recorded the lowest values. These results showed that the *T. harzianum* can serve as a viable eco-friendly alternative to chemical fungicides in integrated disease and crop management.

Keywords: *Bioagents, Groundnut, M phaseolina and S rolfsii*

Groundnut or peanut (*Arachis hypogaea* L.) is an important oil seed crop, grown throughout the tropics. It is the world's 13th most significant food crop and the 4th most important oilseed crop. It is the third most important source of vegetable protein in the world. It originated from South America and belongs to the family Leguminosae. Groundnut was introduced to South-Western India in the 16th century by Portuguese (Thamaraikannan *et al*., 2009). It is generally distributed in the tropical, sub-tropical and warm temperate zones of the world in over 100 countries. Groundnut, as a leguminous plant, has the potential to fix atmospheric nitrogen into the soil, enrich soil and benefits the crop in the rotation.

Globally, China leads in groundnut production with 19.27 million tonnes, followed by India with 10.30 million tonnes (FAO STAT, 2024). In India, the key groundnut producing states include Gujarat, Rajasthan, Andhra Pradesh, Karnataka and Tamil Nadu. In India, about 48.80 lakh hectares were reported under groundnut during 2023-24, compared to 51.21 lakh hectares during the same period in 2022-23. The states of Gujarat (16.35 lakh ha), Rajasthan (8.96 lakh ha), Madhya Pradesh (5.4 lakh ha), Karnataka (4.25 lakh ha) and Andhra Pradesh (3.66 lakh ha) are the major producers of groundnut in India (ANGRAU-CARP, 2023- 2024).

MATERIAL AND METHODS

The paper towel method (Srivastava *et al*., 2002) was used to assess the effect of bioagents on groundnut seed germination and growth. Surface-sterilized seeds were treated with *Trichoderma harzianum*, *Pseudomonas fluorescens* and *Bacillus subtilis* (each @ 1×10x CFU/ ml). A chemical control (tebuconazole @ 1.0%) and an untreated check (pathogen only) were included. Treated seeds were placed on moist blotters, rolled, and incubated at 25/ °C and 80% RH for 14 days.

Total seeds per paper towel- 10 seeds

Design – CRD

Treatments-5

Replications- 4

Germination percentage, shoot length, and root length were recorded and the vigor index was calculated using the formula.:

Vigor index =

(Mean root + shoot length) × Germination (%)

(Abdul-Baki and Anderson, 1973)

RESULTS AND DISCUSSION

Effect of bioagents in promoting plant growth parameters in groundnut

In the present investigation, an attempt was made to observe whether the treatments imposed have any stimulatory (or) inhibitory effect on mean shoot length, root length and germination of groundnut seeds.

Based on the data presented in Table 1, the highest germination percentage was recorded in T_1 (seed treatment with *T. harzianum* at 1 × 10x CFU/ ml), which resulted in 98.5 per cent germination and was on par with T_3 (seed treatment with *B. subtilis* at 1 × 10x CFU/ ml) with 97.0 %. The lowest germination percentage was observed in the untreated control (T_5), which recorded 89.0 %. However, the seed treatment with *P. fluorescens* (T_2) resulted in 91.0 % germination, which was on par with chemical control treatment T_4 (seed treatment with tebuconazole @ 1.0 %), where 94.0 % germination was observed.

A similar trend was observed for shoot length where the maximum shoot length was recorded in T_1 (seed treatment with *T. harzianum* @1 × 10x CFU/ ml) (18.97 cm), followed by T_3 (seed treatment with *B. subtilis* @1 × 10x CFU/ ml) (17.42 cm), T_4 (seed treatment with tebuconazole @1.0 %) (16.05 cm) and T_2 (seed treatment with *P. fluorescens* @1x 10^8 CFU/ ml) (15.22 cm). The minimum shoot length was observed in T_5 (Control) (5.5 cm).

The treatments responded in the same manner for root length where T_1 (seed treatment with *T. harzianum* @1 × 10x CFU/ ml) also recorded the highest value (16.50 cm) and was on par with T_3 (seed treatment with *B. subtilis* @ 1 × 10x CFU/ ml) (16.47 cm), T_4 (seed treatment with tebuconazole @1.0 %) (16.37 cm) and T_2 (seed treatment with *P. fluorescens* @1x 10^8 CFU/ ml) (13.22 cm). The lowest root length was observed in T_5 (Control)(6.57 cm).

The vigor index was significantly high in T_1 (seed treatment with *T. harzianum* @ 1 × 10x CFU/ ml) (3495.20), followed by T_3 (seed treatment with *B. subtilis* @ 1 × 10x CFU/ ml) (3288.60), T_4 (seed treatment with tebuconazole @1.0 %) (3046.80) and T_2 (seed treatment with *P. fluorescens* @1x 10^8 CFU/ ml) (2588.10). The lowest vigor index was recorded in T_5 (Control) (1076.20). All the treatments were significantly superior over control except in germination per cent where seed treatment with *P. fluorescens* was found on par with control.

From these observations, it can be concluded that T_1 (seed treatment with *T. harzianum* @ 1 × 10x CFU/ ml) was the most effective in enhancing plant growth parameters, while the untreated control (T_5) consistently showed the lowest performance across all measured criteria.

Effect of bioagents in promoting plant growth in *S. rolfsii* pretreated seeds of groundnut

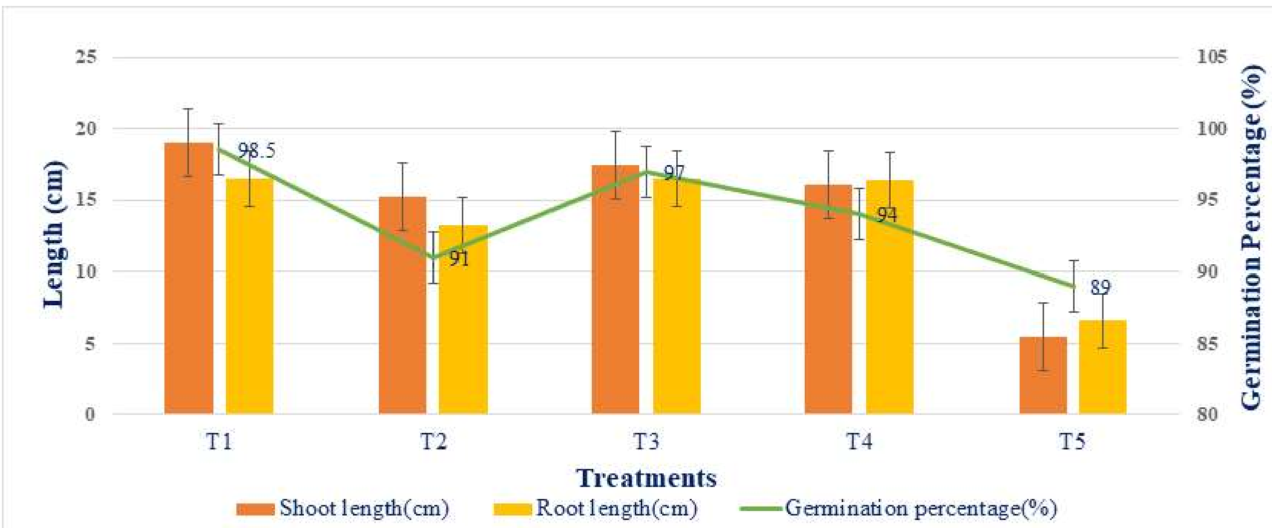
In the present investigation, an attempt was made to observe whether the treatments imposed have any stimulatory (or) inhibitory effect on mean shoot length, root length and germination of *S. rolfsii* pretreated seeds of groundnut.

Based on the data in Table 2, the highest germination percentage was observed in the chemical control treatment T_4 (seed treatment with tebuconazole @ 1.0 %), which was recorded with 86.0 %, followed by T_1 (seed treatment with *T. harzianum* @ 1 × 10x CFU/ ml) with 81.0 %. The lowest germination percentage was observed in the untreated control (T_5), which recorded 61.0 % and was found significantly inferior over other treatments *i.e.*, T_3 (seed treatment with *B. subtilis* @1 × 10x CFU/ ml) with 78.0 % germination and T_2 (Seed treatment with *P. fluorescens* (@1x 10^8 CFU/ ml) where germination was 72.0 %.

The longest shoots were recorded in T_4 (15.90 cm). However, among bioagents the root length was found superior in T_1 (seed treatment with *T. harzianum* @1 × 10x CFU/ ml) (15.65 cm) followed by T_3 (seed treatment with *B. subtilis* @1 × 10x CFU/ ml) (14.00 cm) and T_2 (seed treatment with *P. fluorescens* @1x 10^8 CFU/ ml) (12.00 cm). The shortest shoot length was observed in the control (T_5), measuring 4.95 cm which was found significantly low.

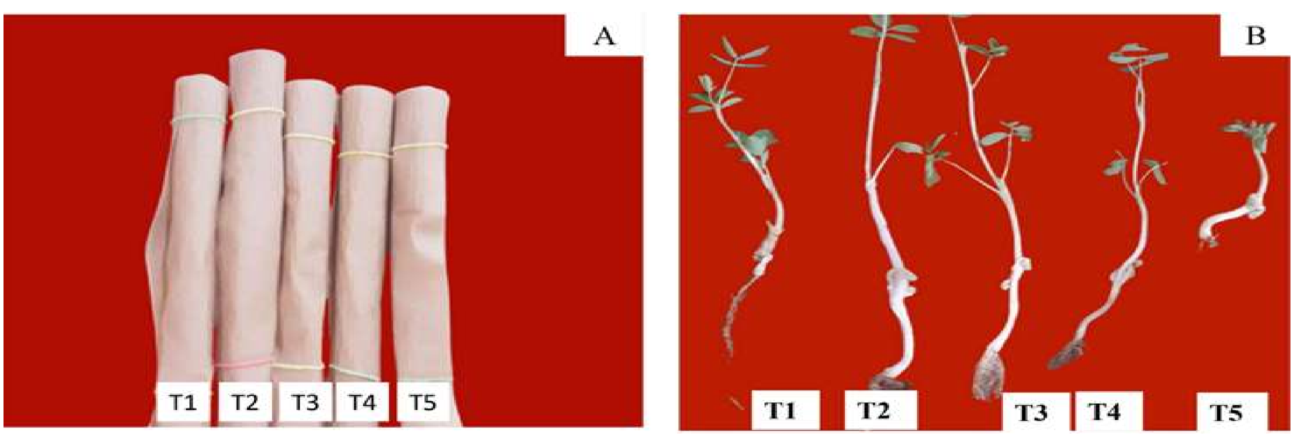
Table 1. Effect of bioagents in promoting plant growth

T.No	Treatments	*Germination per cent (%)	*Shoot length (cm)	*Root length (cm)	*Seedling length (cm)	*Vigor index
1	Seed treatment with *T. harzianum* *(@1x 108 CFU/ ml)*	98.5	18.97	16.5	35.47	3495.2
		-9.97				
2	Seed treatment with *P. fluorescens* *(@1x 108 CFU/ ml)*	91	15.22	13.22	28.45	2588.1
		-9.59				
3	Seed treatment with *B. subtilis* *(@1x 108 CFU/ ml)*	97	17.42	16.47	33.9	3288.6
		-9.89				
4	Seed treatment with tebuconazole @1.0 %	94	16.05	16.37	32.42	3046.8
		-9.74				
5	Control	89	5.5	6.57	12.07	1076.2
		-9.48				
SEm±		0.05	0.35	0.32	0.47	
C.D(P ≤0.05)		0.16	1.06	0.99	1.43	
C.V. (%)		1.08	4.79	4.71	3.3	



T_1= Seed treatment with *T. harzianum* (@1x 10^8 CFU/ ml); T_2= Seed treatment with *P. fluorescens* (@1x 10^8 CFU/ ml) T_3= Seed treatment with *B. subtilis* (@1x 10^8 CFU/ ml); T_4= Seed treatment with tebuconazole @ 1.0 %; T_5 = Control (Distilled water)

Fig 1 Effect of bioagents on plant growth parameters in groundnut



T_1= Seed treatment with *T. harzianum* (@1x 10^8 CFU/ ml); T_2= Seed treatment with *P. fluorescens* (@1x 10^8 CFU/ ml) T_3= Seed treatment with *B. subtilis* (@1x 10^8 CFU/ ml); T_4= Seed treatment with tebuconazole @ 1.0 %; T_5 = Control (Distilled water)

Plate 1 A and B-Growth of groundnut seeds in paper towel assay

Root length was also recorded to be highest in T_4 (seed treatment with tebuconazole @ 1.0 %) (15.17 cm) but was followed by T_1 (seed treatment with *T. harzianum* @1 × 10x CFU/ ml) (14.42 cm), T_3 (seed treatment with *B. subtilis* @ 1 × 10x CFU/ ml) (12.62 cm) and T_2 (Seed treatment with *P. fluorescens* @1x 10^8 CFU/ ml) (10.30 cm) which happened to be bioagents. The lowest root length was observed in T_5 (Control) (3.55 cm).

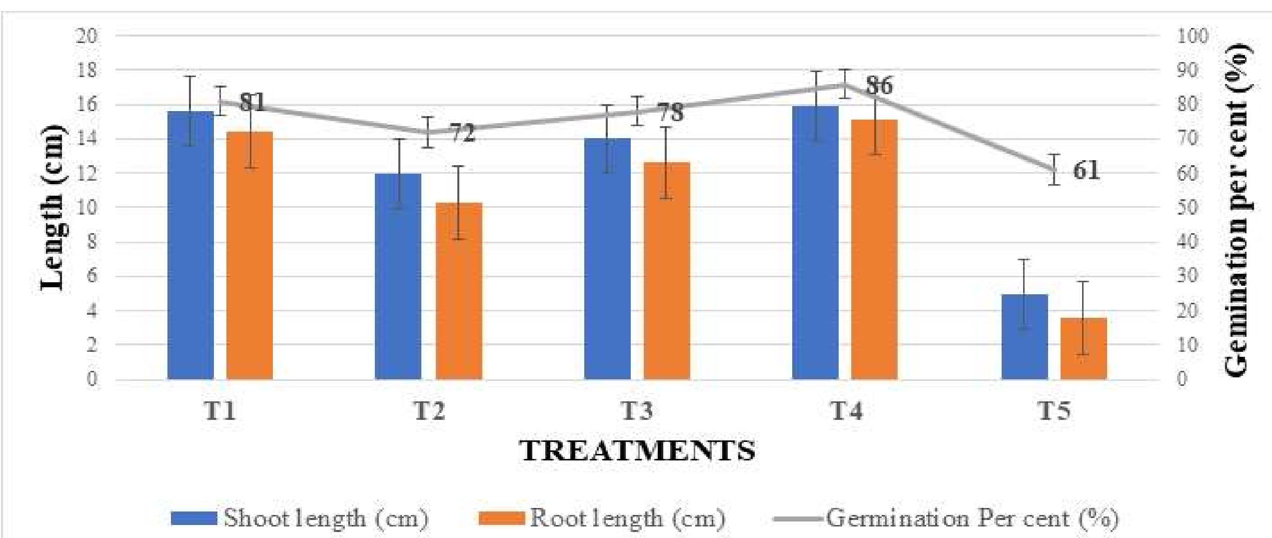
The vigor index was also the highest in T_4 (seed treatment with tebuconazole @ 1.0 %) (2672.45), followed by T_1 (seed treatment with *T. harzianum* @1 × 10x CFU/ ml) (2436.08), T3 (seed treatment with *B. subtilis* @1 × 10x CFU/ ml) (2076.75) and T_2 (seed treatment with *P. fluorescens* @1x 10^8 CFU/ ml) (1605.60). The lowest vigor index was recorded in the untreated control (T_5), which was 518.50.

From these observations, it can be concluded that among all treatments, the chemical control (T_4) showed the best performance across all growth parameters including germination percentage, shoot

Table 2. Effect of bioagents in promoting plant growth in *S. rolfsii* pretreated seeds of groundnut

T.No	Treatments	*Germination per cent (%)	*Shoot length (cm)	*Root length (cm)	*Seedling length (cm)	*Vigor index
1	Seed treatment with *T. harzianum*	81	15.65	14.42	30.07	2436.08
	(@1x 10^8 CFU/ ml)	-9.05				
2	Seed treatment with *P.*	72	12	10.3	22.3	1605.6
	(@1x 10^8 CFU/ ml)	-8.54				
3	Seed treatment with *B. subtilis*	78	14	12.62	26.62	2076.75
	(@1x 10^8 CFU/ ml)	-8.87				
4	Seed treatment with Tebuconazole	86	15.9	15.17	31.07	2672.45
	@ 1.0 %	-9.32				
5	Control	61	4.95	3.55	8.5	518.5
		-7.87				
SEm±		0.09	0.28	0.2	0.29	
C.D(P ≤0.05)		0.28	0.85	0.61	0.9	
C.V. (%)		2.13	4.47	3.59	2.5	

Mean of four replications, Figures in parenthesis are square root transformed values



T_1= Seed treatment with *T. harzianum* (@1x 10^8 CFU/ ml); T_2= Seed treatment with *P. fluorescens* (@1x 10^8 CFU/ ml) T_3= Seed treatment with *B. subtilis* (@1x 10^8 CFU/ ml); T_4= Seed treatment with tebuconazole @ 1.0 %; T_5 = Control (Pathogen control)

Fig. 2. Effect of bioagents in promoting plant growth in *S. rolfsii* pretreated seeds of groundnut

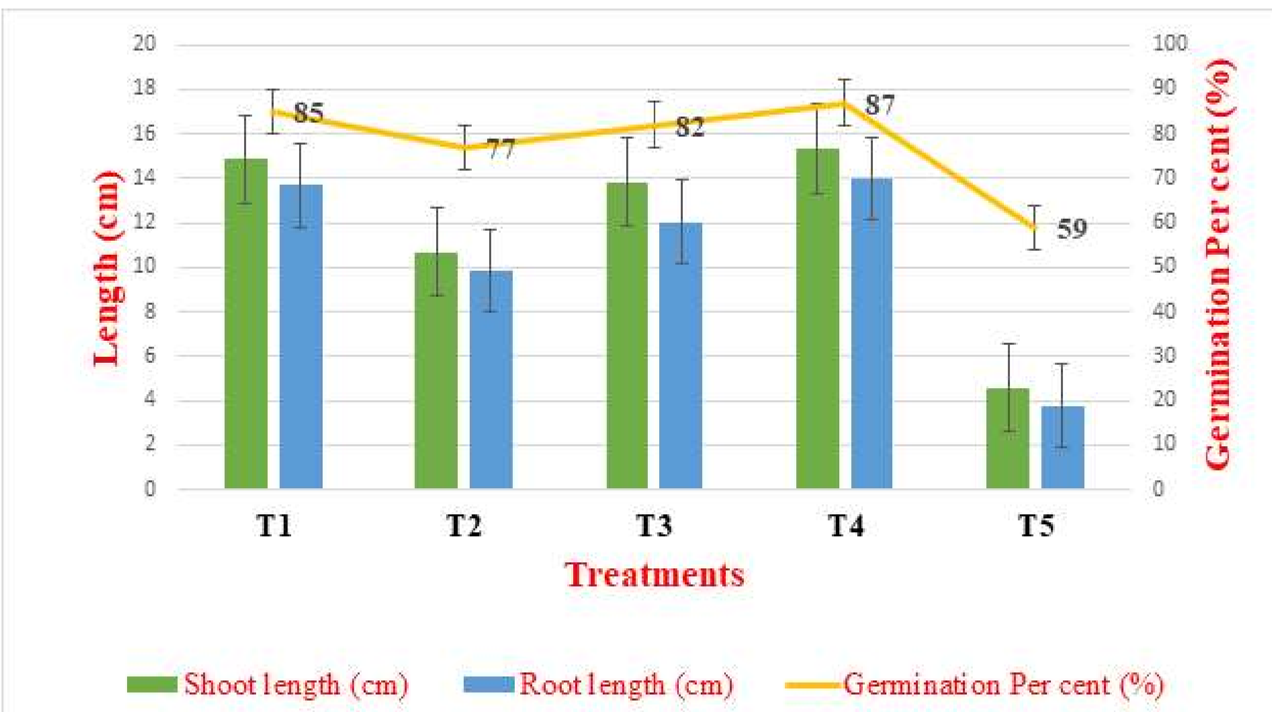
(A) T_1-Seed treatment with *T. harzianum* (@1x 10^8 CFU/ ml); (B) T_2-Seed treatment with *P. fluorescens* (@1x 10^8 CFU/ ml) (C) T_3-Seed treatment with *B. subtilis* (@1x 10^8 CFU/ ml); (D) T_4-Seed treatment with tebuconazole @1.0% (E) T_5-Control (Pathogen control)

Plate 2. Effect of bioagents on the growth parameters of groundnut seeds pretreated with *S. rolfsii* in paper towel assay(a-e)

Table 3. Effect of bioagents in promoting plant growth in *M. phaseolina* pretreated seeds of groundnut

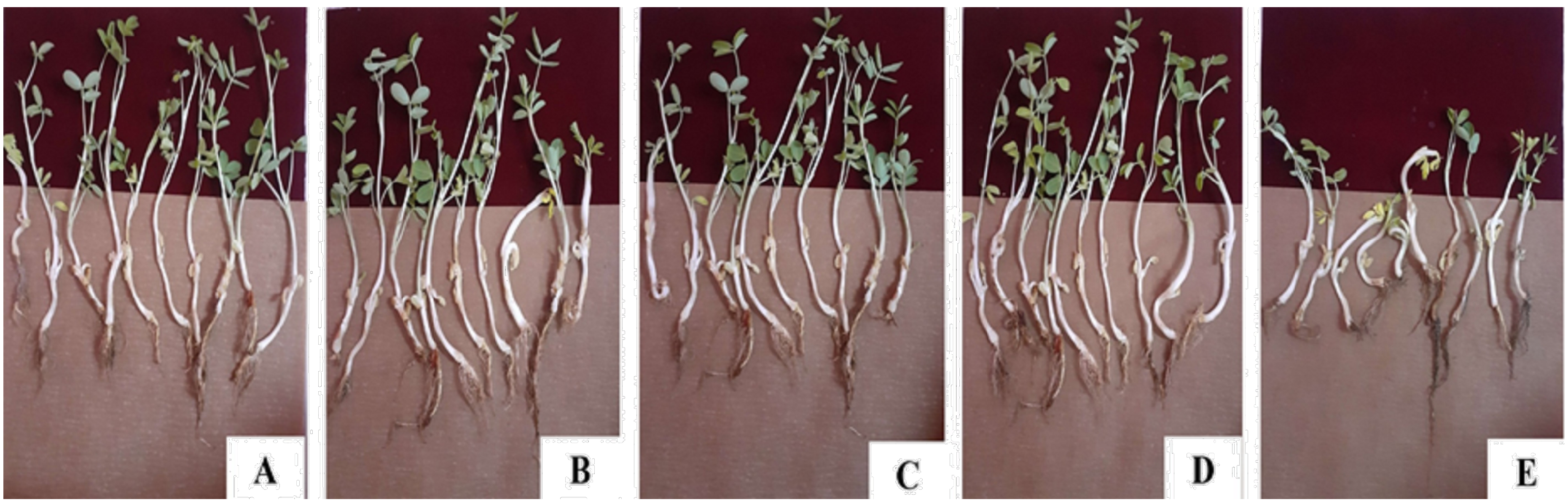
T. No	Treatments	*Germination per cent (%)	*Shoot length (cm)	*Root length (cm)	*Seedling length (cm)	*Vigor index
1	Seed treatment with *T. harzianum* (@1x 10^8 CFU/ ml)	85 -9.27	14.85	13.67	28.52	2424.62
2	Seed treatment with *P. fluorescens* (@1x 10^8 CFU/ ml)	77 -8.83	10.7	9.85	20.55	1582.35
3	Seed treatment with *B. subtilis* (@1x 10^8 CFU/ ml)	82 -9.11	13.85	12.05	25.9	2123.8
4	Seed treatment with tebuconazole @1.0 %	87 -9.37	15.32	13.97	29.3	2549.1
5	Control	59 -7.74	4.57	3.77	8.35	492.65
SEm±		0.09	0.18	0.17	0.24	
C.D(P ≤0.05)		0.27	0.56	0.53	0.75	
C.V. (%)		2.04	3.12	3.26	2.2	

Mean of four replications, Figures in parenthesis are square root transformed values



T_1= Seed treatment with *T. harzianum*. (@1x 10^8 CFU/ ml); T_2= Seed treatment with *P. fluorescens* (@1x 10^8 CFU/ ml) T_3= Seed treatment with *B. subtilis* (@1x 10^8 CFU/ ml); T_4= Seed treatment with tebuconazole @ 1.0 %; T_5 = Control (Pathogen control)

Fig. 3. Effect of bioagents in promoting plant growth in *M. phaseolina* pretreated seeds of groundnut



A) T_1-Seed treatment with *T. harzianum* (@1x 10^8 CFU/ ml);
B) T_2-Seed treatment with *P. fluorescens* (@1x 10^8 CFU/ ml)
C) T_3-Seed treatment with *B. subtilis* (@1x 10^8 CFU/ ml);
D) T_4-Seed treatment with tebuconazole @1.0 %;
E) T_5-Control (Pathogen control)

Plate. 3. Effect of bioagents on the growth parameters of groundnut seeds pretreated with *M. phaseolina* seeds in paper towel assay(A-E)

and root lengths, and vigor index. However, T_1 (seed treatment with *T. harzianum* @1 × 10x CFU/ ml) was the most effective among the bioagents, showing results closely comparable to the chemical control. The untreated control (T_5) consistently recorded the lowest values, indicating the importance of seed treatment in enhancing early plant growth.

CONCLUSION

The effect of bioagents on plant growth parameters like germination per cent, seedling length and vigor index were evaluated by adopting paper towel assay method under both healthy and pathogen inoculated conditions. The chemical control treatment (tebuconazole @ 1.0%) consistently exhibited superior performance across all measured parameters, achieving the highest germination percentage (87.0%), shoot length (15.32 cm), root length (13.97 cm) and vigor index (2549.10).

Among the bioagents, *Trichoderma harzianum* (T_1) emerged as the most effective, with results closely comparable to those of the chemical treatment. It recorded 85.0% germination, 14.85 cm shoot length, 13.67 cm root length and a vigor index of 2424.62. This suggests that *T. harzianum* possesses strong growth-promoting and bioprotective capabilities, enabling it to perform effectively even in the presence of soilborne pathogens such as *Sclerotium rolfsii* and *Macrophomina phaseolina*.

Given its consistent performance and ability to suppress pathogen effects, *T. harzianum* can be viewed as a viable and sustainable seed treatment option in integrated disease management programs. Its use could support early crop establishment, boost plant vigor, and help reduce the environmental pollution linked to chemical fungicide use. This approach fits well with the goals of sustainable and climate-resilient agriculture.

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