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RESEARCH ARTICLE

Unshackle *Pseudomonas putida* GN1 and organic amendments against root rot (*Macrophomina phaseolina*) and stem rot (*Sclerotium rolfsii*) control by plant growth promotion in groundnut

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Abstract

The efficacy of *Pseudomonas putida* (GN1) and *Burholderia cepacia* (KKM1) as seed treatment and soil application along with neem cake against groundnut root rot and stem rot disease was evaluated under rainfed conditions during the years 2021, 2022 and 2023. The present study is to evaluate biocontrol agents and organic amendments against *Macrophomina phaseolina* and *Sclerotium rolfsii* in peanut. The result showed that neem cake (10 %) recorded the maximum inhibition of *Macrophomona phaseolina* (40.91 %) and *S. rolfsii* (45.45 %) under *in vitro* whereas neem cake combined with *P. putida* and *B. cepacia* as seed treatment 10 g/kg of seed and soil Application of *P. putida* @ 2.5 kg + Neem cake 150 kg/ha decreases the soil-borne diseases of root rot (71.70 %) and stem rot (64.88 %) and also increased the yield 2130.48 kg/ha with the cost-benefit ratio of 2.74 under rainfed field conditions comparing to other treatments. In addition, the same treatment increased the total root length (2110.41 mm), Root tips (573 Nos), forks (501 Nos), Maximum diameter (18.11 mm) and estimated volume (27170 cm^3) when using BioVis PSM Root - Rhizoscanner.

Keywords

Pseudomonas putida; root rot; rooting pattern; stem rot

Introduction

In India, the taste and preference for oilseed consumption varied over time and region. Among the edible oils consumed in rural and urban areas, groundnut oils contribute 59 % of the total consumption. Groundnut crop is grown over an area of 29.59 (mha) over the world, with a total production of 48.75 million tonnes (MT) (1). In India, groundnut is grown in an area of 4.8 mha with a production of 9.9 MT and an average productivity of 2.06 tonnes ha^{-1} (2). About 72 % of the area of oilseeds falls under rainfed farming where biotic threats (diseases) and climate vagaries cause severe damage to crops. Soil-borne diseases incited by *Aspergillus niger*, *Sclerotium rolfsii* and *Rhizoctonia bataticola* were the key constraints attributed to groundnut production (3). The treatment module consists of soil application with *T. harzianum* (Th-BKN) @ 10 kg ha^{-1} + FYM @ 10 tonnes ha^{-1} gave the highest root length, shoot length, fresh weight, dry weight, disease control, pod yield in kg ha^{-1} and lowest disease incidence (9.90 cm, 21.05 cm, 222.60 g, 65.20 g, 86.30 %, 2173 kg ha^{-1} and 7.51 %) (4). Among twenty isolates the *P. fluorescens* isolates Pf4-99, were found effective in controlling root rot pathogen and promoting plant growth in chickpeas (5). This is particularly true if applied antagonists can proliferate sufficiently to colonize the germinating seeds and the developing roots (6). A phytopathogenic fungus, *Macrophomina phaseolina*, which infects a wide range of plants, a jute endophytic bacterium, *Burkholderia contaminans* NZ, was found to have a promising effect in controlling the fungus *in vitro* culture conditions using the iTRAQ LC-MS/MS method for quantitative proteomics study (7). *Burkholderia cepacia* strains are naturally present in large numbers associated with corn roots. When used in seed treatment trials, these strains have the potential to colonize roots and produce antibiotics to protect against pathogens, the main objectives for oilcakes have been less explored in research so far hence the present study aims to evaluate biocontrol agents and organic amendments against management of root rot and stem rot of groundnut.

Materials and Methods

In vitro evaluation

Preparation of aqueous extracts of oilcake

The required quantity of oil cakes (five oil cakes viz, pungam cake, Gingelly cake, Castor cake, Mahua cake and neem cake and FYM, RHA, decomposed coir pith and biochare) was taken and made into powder. It was soaked in sterile distilled water (1 g in 1.25 mL). The material was ground using a pestle and mortar and filtered through a muslin cloth and the filtrate was centrifuged at 10000 rpm for 15 min. The supernatant served as the standard oil cake extract solution (100 %) (8).

Testing the antifungal activity of extracts of oil cake in vitro against Macrophomina phaseolina and S. rolfsii

The efficacy of oil cake extracts was tested against *S. rolfsii* and *M. phaseolina* using the poisoned food technique. The freshly prepared Potato Dextrose Agar medium was distributed to several conical flasks @ 50 mL per conical flask. Aqueous extracts of oil cake @ 5 mL were mixed with 50 mL of Potato Dextrose Agar medium to obtain 10% and sterilized. The sterilized Potato Dextrose Agar medium was poured on sterilized Petri plates @ 15 mL per petri plate and then allowed to solidify. Mycelial disc (9 mm) of *Pasolini* and *S. rolfsii* was taken from the actively growing culture and placed at the centre of each Petri plate and incubated at room temperature. The Potato Dextrose Agar medium without any extracts of oil cake served as control. The radial growth (cm) of *S. rolfsii* was recorded after three days of incubation. Sclerotial production was observed at 15 days after incubation.

Compatibility of oil cakes with biocontrol agents in solid medium (In vitro)

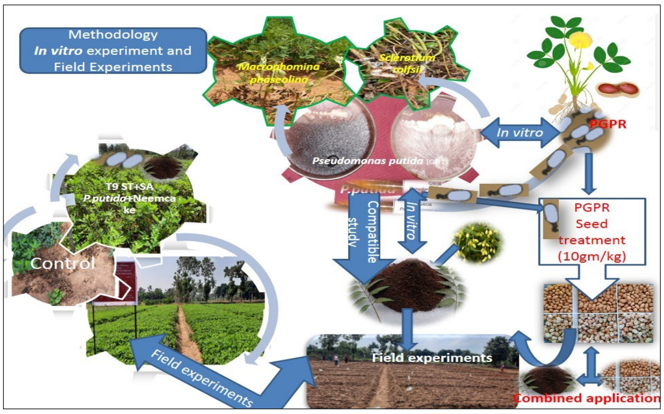
The growth of antagonists in extracts of oil cake was tested following poisoned food technique. The 10 % concentration of oil cake extracts was prepared and tested for their compatibility with *Trichoderma asperellum*, *Pseudomonas putida* (GN1), *B. cepacia* (KKM1) and *Pseudomonas Sp* (K1) and *B. subtilis* (Bbv54). The Petri plates containing PDA medium and King's B medium were inoculated with a 9 mm disc of Trichoderma and streaked with *P. putida, B. cepacia*, *Pseudomonas* spp and *B. subtilis* respectively. They were incubated at room temperature. The mycelial diameter of *T. asperellum* and the growth of *P. putida, B. cepacia* and *Pseudomonas* spp were recorded after 3 days of incubation.

Field experiments

Plant growth-promoting rhizobacteria (PGPR) isolated from groundnut rhizosphere were tested under *in vitro* condition. Two PGPRs were effective against root rot and stem rot pathogens. These were molecularly characterized and identified as *P. putida* GN1 (ON307464) and *B. cepacia* KKM 1 (OM908755).

These PGPR, *T. asperellum*, *B. subtilis* individual and combined with organic amendments for Neem cake and coirpith compost for tested for three consecutive years 2021, 2022 and 2023 with different agro climatic locations. The methods of application for seed treatment alone and seed treatment-cum-soil application with Random block design and three replications in hotspot area of Tamil Nadu. The experiment was conducted with a susceptible variety VRI 2 (Table S1).

Germination count was taken at 7 Days after sowing. The yield parameters were recorded and analysed statistically. The percentages of disease incidence were observed at regular intervals. In addition, seed yield (kg/ha), was also recorded.



Plant growth promotion

Groundnut plants for alteration of rooting pattern study after harvesting, three plants were selected from each treatment and washed with tap water and studied the different root characteristics using Biovis software rhizoscanner (DARS, Chettinad, TNAU, Coimbatore) analysis.

Statistical analysis

The collected data underwent statistical analysis using Microsoft Excel for Windows 2007. Subsequently, Duncan's multiple range test (DMRT) was performed at a significance level of 5 %. Prior to ANOVA, the percentage values of the disease index were transformed using an arcsine function. The data were subjected to ANOVA at significance level ($P < 0.05$), followed by the application of Duncan's Multiple Range Test (DMRT) to compare means among the groups.

Results

Among the different organic amendments tested, the neem cake extract (10 %) effectively controlled *M. phaseolina* (40.91%) and *S. rolfsii* (45.95 %) as compared to other treatments followed by mahua cake extract (10 %) recorded 22.73% and 26.14% reduction over control respectively (Table 1).

These neem cake, mahua cake, Farmyard manure and decomposed coir pith showed strong compatibility study with *P. putida* and *B. cepacia* (Table 2).

Field experiments

Root rot

Among the treatments, three-year data showed that the lowest root rot disease incidence (8.73 %) was recorded in the treatment viz., T9 Seed treatment 10g/kg seed + Soil application *P. putida* (GN1) @ 2.5 kg ha^{-1} + Neem cake 150 kg ha $^{-1}$ and showed on par with propiconozole (8.77%). Followed by the treatment T8 ST (*P. putida*) 10 g/kg + SA (*P. putida*) @2.5 kg ha^{-1} + Composted Coir pith @ 5T ha^{-1} and T1 ST (*P. putida* (GN1)) + SA (*P. putida* (GN1)) @ 2.5 kg ha^{-1} recorded the less root rot disease incidence (10.04% and 10.61%) which on par with each other (Table 3).

Stem rot

Among the treatments in the three years data, the lowest stem rot disease incidence (9.63 %) was recorded in the treatment propiconozole which is on par with the treatment (T9): seed treatment 10g/kg seed + Soil application of *P. putida* @2.5kg + Neem cake 150kg ha^{-1} and on par with (10.04 %) the treatment (T1) ST (*P. putida* (GN1)) + SA (*P. putida* (GN1)) @ 2.5 kgha^{-1} and (T8) ST (*P. putida* (GN1) 10g/kg +SA (*P. putida* (GN1)) @ 2.5 kg ha^{-1} + composted coir pith @ 5Tha^{-1} and recorded the less root rot disease incidence (10.65 % and 10.79 %) (Table 3a).

Table 2. Compatibility of oil cakes with bio control agents in solid medium (*in vitro*)

S. No	Oil cakes (10%)	Growth of *P.putida* (GB08)	Growth of *Burholderia*
1	Pungam cake	+	+
2	Gingelly cake	+	+
3	Castor cake	+	+
4	Neem cake	++	++
5	Mahua cake	++	++
6	FYM	++	++
7	RHA	+	+
8.	Decomposed coirpith	++	++
9.	Biochare	+	+
10.	Control	++	++

+ Low growth; + + full growth; * extracts at 10 % concentration

Table 1. Effect of Organic amendments against *Macrophomina phaseolina* and *Sclerotium rolfsii*

S. No	Organic amendments (10%)	Mycelial growth of *Macrophomina phaseolina* (cm)	Percent reduction over control (%)	Mycelial growth of *S.rolfsii* (cm)	Percent reduction over control (%)
1.	Pungamcake	7.00	20.45	6.80	23.93
2.	Gingelly cake	8.20	6.82	7.50	14.77
3.	Castor cake	7.80	11.36	6.90	21.59
4.	Neem cake	5.20	40.91	4.80	45.45
5.	Mahua cake	6.80	22.73	6.50	26.14
6.	FYM	7.20	18.18	6.60	25.00
7.	RHA	8.20	6.80	8.00	9.09
8.	Decompostedcoirpith	6.70	23.86	6.60	25.00
9.	Biochare	8.00	9.00	6.70	23.86
10.	Control	8.80		8.80	
	CD (0.05%)	0.86		0.91	

Table 3. Effect of biocontrol and organic amendments against root rot (*Macrophomina phaseolina*) soil-borne disease of rainfed groundnut (Three years)

S. No	Treatments	Germi Nations (%)	Root rot disease incidence (%) 2021	2022	2023	Mean disease incidence (%)
T1	Seed treatment 10g/kg and soil applications of *Pseudomonas putida* @ 2.5kg/ha	95.02	10.26	11.78	9.8	10.61
T2	Seed treatment 10g/kg and soil applications of *Burkholderia cepacia* @ 2.5kg/ha	92.56	11.58	12.45	13.5	12.51
T3	Seed treatment 4g/kg and soil applications of *Trichoderma asperellum*@2.5kg/ha	93.36	13.25	15.2	10.28	12.91
T4	Seed treatment 10/kg and soil applications of *Bacillus subtilis* @2.5kg/ha	93.46	12.1	14.46	12.26	12.94
T5	Soil Application (SA) of Neem cake (NC) @150 kg/ha	89.69	15.75	17.78	18.52	17.35
T6	Soil application of Composted coirpith @ 5t/ha	87.62	18.42	20.76	21.2	20.13
T7	Soil application of Composted coirpith @ 5t/ha+ Neem cake @ 150 kg/ha	88.64	14.24	16.44	18.56	16.41
T8	Seed treatment 10g/kg and soil applications of *Pseudomonas putida* @(2.5kg/ha) + Composted Coir pith @5T/ha	95.07	9.67	10.46	10	10.04
T9	Seed treatment 10g/kg and soil applications of *Pseudomonas* putida @2.5kg + Neem cake150kg/ha	95.37	8.21	9.28	8.7	8.73
T10	Propiconazole	89.33	8.2	9.6	8.5	8.77
T11	Untreated control	81.00	27.38	38.5	26.68	30.85
	C.D.					2.418
	SE(m)					0.814
	SE(d)					1.151
	C.V.					6.372

Table 3a. Effect of biocontrol and organic amendments against Stem rot (*Sclerotium rolfsii*) soil-borne disease of rainfed groundnut (Three years data)

S. No	Treatments	Germi Nations (%)	Stem rot disease incidence (%)			Mean disease incidence (%)
			2021	2022	2023	
T1	Seed treatment 10g/kg and soil applications of *Pseudomonas putida* @ 2.5kg/ha	95.02	11.78	8.64	11.54	10.65
T2	Seed treatment 10g/kg and soil applications of *Burkholderia cepacia* @ 2.5kg/ha	92.56	12.51	9.6	13.26	11.79
T3	Seed treatment 4g/kg and soil applications of *Trichoderma asperellum*@2.5kg/ha	93.36	12.6	9.8	12.28	11.56
T4	Seed treatment 10/kg and soil applications of *Bacillus subtilis* @2.5kg/ha	93.46	12.51	9.7	12.88	11.70
T5	Soil Application (SA) of Neem cake (NC) @150 kg/ha	89.69	16.92	12.4	16.28	15.20
T6	Soil application of Composted coirpith @ 5t/ha	87.62	19.44	13.18	20.26	17.63
T7	Soil application of Composted coirpith @ 5t/ha+ Neem cake@ 150 kg/ha	88.64	13.95	10.3	15.62	13.29
T8	Seed treatment 10 g/kg and soil applications of *Pseudomonas putida* @(2.5 kg/ha) + Composted Coir pith @5T/ha	95.07	11.58	8.6	12.2	10.79
T9	Seed treatment 10 g/kg and soil applications of *Pseudomonas putida* @ 2.5 kg + Neem cake 150 kg/ha	95.37	11.51	8.2	10.42	10.04
T10	Propiconazole	89.33	10.28	8.4	10.2	9.63
T11	Untreated control	81.00	32.75	24.3	28.76	28.60
	C.D.	1.46		C.D.		1.988
	SE(m)	0.491		SE(m)		0.669
	SE(d)	0.695		SE(d)		0.947
	C.V.	0.935		C.V.		8.452

Efficacy of Groundnut PGPR against soil-borne disease (Pooled data)

Among the treatments in the three years data revealed that the treatment (T9): Seed treatment 10gm/kg seed + Soil application of *P. putida* (GN1) @2.5 kg + Neem cake 150 kg/ha (71.70 % disease reduction) of root rot and Stem rot (64.88 % reduction) and showed on par with chemical treatment propiconozole (71.58 % disease reduction and 66.34 % reduction) (Table 4).

Yield

Among the treatments, ST (10 g/kg) and SA of *P.* putida (GN1) @ 2.5 kg ha^{-1} + Neem cake 150 kg ha^{-1} received the highest yield 2130.48 kg ha^{-1} with the increasing cost benefit ratio of 1:2.74 under rainfed field conditions compared to other treatments (Table 5).

Alteration of root characters

Total root length

The application of *P. putida* (GN1) (2.5 kg ha^{-1}) + Neem cake 150 kg ha^{-1} soil application noticed a longer root length of 2110.412 mm than the control (400.15 mm) while other PGPR recorded shorter root length 15 days after germination (Table 6 and Fig. 1).

Root tips

Soil application of *P. putida* (GN1) (kg ha^{-1}) + Neem cake 150 kg ha^{-1} as soil application recorded higher root tips of 573 Nos than the control (112 Nos) (Table 6 and Fig. 1).

Forks

Maximum number of root forks (501 nos) was noticed in *P. putida* (GN1) (2.5 kg ha^{-1}) + Neem cake 150 kg ha^{-1} as soil application followed by *B. cepacia* + Neem cake (391 nos) (Table 6 and Fig. 1) other PGPR treated and control plants showed less number of forks.

Table 4. Effect of biocontrol agents and organic amendments against groundnut soil-borne disease reduction (Pooled three year data)

S. No	Treatments	Pooled data analysis (2020-2023)			
		Root rot disease incidence (%)		Stem rot Disease incidence (%)	
		Mean diseases incidence (%)	Percent reduction over control	Mean Diseases incidence (%)	Percent reduction over control
T1	Seed treatment 10g/kg and soil applications of *Pseudomonas putida* @ 2.5kg/ha	10.61	65.60	10.65	62.75
T2	Seed treatment 10g/kg and soil applications of *Burkholderia cepacia* @ 2.5kg/ha	12.51	59.45	11.79	58.78
T3	Seed treatment 4g/kg and soil applications of *Trichoderma asperellum*@2.5kg/ha	12.91	58.15	11.56	59.58
T4	Seed treatment 10/kg and soil applications of *Bacillus subtilis* @2.5kg/ha	12.94	58.06	11.70	59.10
T5	Soil Application (SA) of Neem cake (NC) @150 kg/ha	17.35	43.76	15.20	46.85
T6	Soil application of Composted coirpith @ 5t/ha	20.13	34.76	17.63	38.37
T7	Soil application of Composted coirpith @ 5t/ha+ Neem cake@ 150 kg/ha	16.41	46.80	13.29	53.53
T8	Seed treatment 10g/kg and soil applications of *Pseudomonas putida* @(2.5kg/ha) + Composted Coir pith @5T/ha	10.04	67.44	10.79	62.26
T9	Seed treatment 10g/kg and soil applications of *Pseudomonas* putida @2.5kg + Neem cake150kg/ha	8.73	71.70	10.04	64.88
T10	Propiconazole	8.77	71.58	9.63	66.34
T11	Untreated control	30.85	-	28.60	-

*ST-Seed Treatment, * SA-Soil Applications

Table 5. Effect of biocontrol and organic amendments against soil-borne disease of rainfed groundnut in yield (Three years pooled data)

S. No	Treatments	Yield(kg/ha)			Average Mean Yield (Kg/ha)	BC ratio
		2021	2022	2023		
T1	Seed treatment 10g/kg and soil applications of *Pseudomonas putida* @ 2.5kg/ha	1995.6	1990.5	2200.6	2062.23	1: 2.59
T2	Seed treatment 10g/kg and soil applications of *Burkholderia cepacia* @ 2.5kg/ha	1975	1970.6	1980	1975.20	1: 2.48
T3	Seed treatment 4g/kg and soil applications of *Trichoderma asperellum*@2.5kg/ha	1990.7	1980.8	2100.4	2023.96	1: 2.55
T4	Seed treatment 10/kg and soil applications of *Bacillus subtilis* @2.5kg/ha	1985.6	1975.6	2148.4	2036.53	1: 2.56
T5	Soil Application (SA) of Neem cake (NC) @150 kg/ha	1890.68	1870.8	1896.5	1885.99	1: 2.35
T6	Soil application of Composted coirpith @ 5t/ha	1820.25	1780.4	1760.8	1787.15	1: 2.22
T7	Soil application of Composted coirpith @ 5t/ha+ Neem cake@ 150 kg/ha	1870.75	1840.5	1920.78	1877.34	1: 2.30
T8	Seed treatment 10g/kg and soil applications of *Pseudomonas putida* @ (2.5kg/ha) + Composted Coir pith @5T/ha	2010	2000	2250	2086.66	1:2.58
T9	Seed treatment 10g/kg and soil applications of *Pseudomonas* putida @2.5kg + Neem cake150kg/ha	2040.25	2010.4	2340.8	2130.48	1: 2.74
T10	Propiconazole	2028	2060.8	2020	2036.26	1: 2.55
T11	Untreated control	1690	1540	1670	1633.33	1:2.00
			C.D.		122.569	
			SE(m)		41.258	
			SE(d)		58.348	
			C.V.		3.65	

Root diameter

Maximum Root diameter was observed in *P. putida* (GN1) (2.5 kg ha^{-1}) + Neem cake 150 kg ha^{-1} as soil application (18.11 mm) followed by *Trichoderma asperellum* + Neem cake (15.07 mm) (Table 6 and Fig. 1) other PGPR treated and control plants showed less root diameter.

Root volume

The root volume was found to be higher in *P. putida* (GN1) (kg ha^{-1}) + Neem cake 150 kg ha^{-1} as soil application (27170 cum^2) treated plants (Table 6 and Fig. 1), other PGPR recorded less root volume.

Table 6. Alteration of rooting architecture by *Pseudomonas putida* (GN1) in groundnut

Treatments	Total length (mm)	Root tips (Nos)	Forks (Nos)	Segments (Nos)	Dia (min) mm	Dia (max) mm	Dia (av) mm	Est root volume (Cumm)
Pseudomonas putida + Neem cake	2110.41[a]	573[a]	501[a]	1055[a]	1693[b]	18.11[a]	2.59[f]	21170.46[a]
Burholderia cepacia + Neem cake	1712.82[b]	199[d]	391[b]	454[b]	1693[b]	13.54[d]	2.96[c]	19100.61[b]
Trichoderma asperellum + Neem cake	1571.97[c]	205[c]	317[c]	422[c]	3387[a]	15.07[b]	2.75[e]	14059.91[c]
Neem cake	1388.15[d]	221[b]	230[d]	359[d]	1693[b]	14.39[c]	2.85[d]	13664.89[e]
Propiconazole	953.87[e]	189[e]	222[e]	290[e]	1693[b]	13.20[e]	3.40[a]	14045.95[d]
Control	400.15[f]	112[f]	83[f]	133[f]	3387[a]	11.17[f]	3.06[b]	3994.95[f]

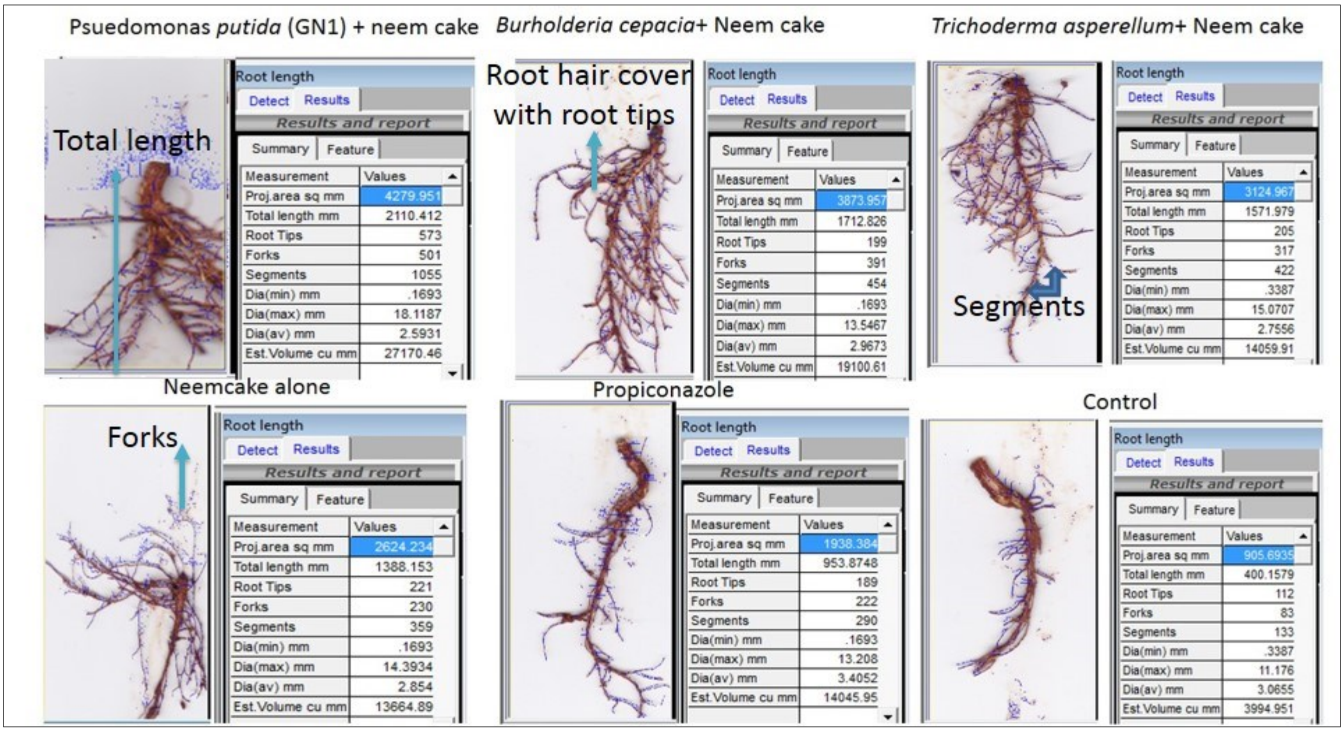


Fig. 1. Alteration of rooting character by *Pseudomonas putida* (GN1) in groundnut.

Discussion

A significant shift in the soil fungal community structure is associated with each organic material application, organic materials frequently improve soil fertility and structure by enhancing soil organic matter contents and nutrient status, thereby increasing soil microbial biomass and activity. Our results indicated that neem cake (10 %) showed reduction of *M. phaseolina* (40.91 %) and *Sclerotium rolfsii* (45.45%) pathogens in groundnut *in vitro*. Similarly, the oilcakes of Neem, Mahua, Gingelly, Groundnut and Coconut were evaluated against Fusarium. Among the five oil cakes tested, Neem cake recorded the inhibition of 18.66 % and 55.55 % at 5 % and 10 % concentrations against fusarium under in vitro conditions, followed by mahua cake (16.22 % and 51.33 %) (9).

Similarly, the castor cake @ 500 kg ha^{-1} along with *T. harzianum* @ 1.5 kg ha^{-1} at the time of sowing was found to be most effective for stem rot infection in groundnut with highest pod yield of 2148 kg ha^{-1} (10). Organic amendments induce the association of beneficial microflora around the rhizosphere, which can help to reduce the plant pathogens in the soil (11). The *P.* spp. EGN 1 had great potential as a bio-stimulant and biocontrol agent to manage stem rot in peanuts effectively (12).

The combined application of the biocontrol agent with an organic amendment is a biological approach to control soilborne diseases in the present study. The seed treatment of 10 g/kg seed + Soil application of *P. putida* @ 2.5 kg + Neem cake 150 kg ha^{-1} recorded less incidence of root rot and stem rot which is on par with the chemical fungicide propiconozole . These results supported organic amendments to suppress soil borne pathogens (13). Several studies also suggest that when bacterial or fungal antagonists such as *P. flourescens* or *Trichoderma* spp. were used in combination with organic amendments, their antagonistic efficacy was enhanced (14). Greenhouse experiments showed that in the presence of pathogens, all antagonists increased the growth indices of soybean in both pasteurized and non-pasteurized soil. Reductions of microsclerotia coverage on soybean root and stem by *P. agglomerans*, *Bacillus* sp. and *T. harzianum* were up to 62.5, 87.6 and 62.5 %, respectively and for maneb fungicide it was 87.6 % in pasteurized soil. The highest reduction in pre-emergence (97.61 %), post-emergence (95.77 %) and average (96.69 %) mortality were recorded with the treatment of thiram + carbendazim + *T. harzianum* + *P. fluorescens* + NSC + *A. indica* extract against groundnut stem rot and pod rot (15).

P. putida mechanisms such as phytohormone synthesis, nutrient solubilization, adaptation to different stress conditions and excellent root colonization ability (16). The combined application of *P. putida* along with neem cake showed increasing root diameter and root length which helps in absorbing more water from soil under deficit conditions. Further, increasing root hairs, root tips, forks and root volume to encourage groundnut growth under drought condition. *P. putida* KT2440 triggered ISR response against *Colletotrichum graminicola* in corn (17). *P. putida* strains jointly with AM fungi showed antagonistic potential against soil borne pathogens, such as *F. oxysporum*, *Ceratocystis fimbriata* and *Sclerotium rolfsii* (18). Siderophore Pyoverdine, capable of colonizing plant roots, has been shown to facilitate iron uptake by plants in different model systems (19) produced by promising biocontrol products formulated with *P. putida* (20, 21). The aforementioned works represent a good approach for the development of a *P. putida*-based product for its application in agriculture. Inoculation of rice plants with *P. putida* KT2440 stimulates an alternative plant defense mechanism based on abscisic acid accumulation (22). In the absence of pathogen, *P. putida* reduced ethylene (ET) production and promoted root and stem elongation. Interestingly, gene *OsHDA702*, which plays an important role in root formation (22) by *P. putida* KT2440 where three T6SS gene clusters (K1-, K2- and K3-T6SS) have been identified. Besides, 10 T6SS effector-immunity pairs were found, including putative nucleases and pore-forming colicins. The K1-T6SS is a potent antibacterial device, which secretes a toxic Rhs-type effector Tke2. Remarkably, *P. putida* eradicates a broad range of bacteria in a K1-T6SS-dependent manner, including resilient phytopathogens (23). Similar results were found that seed treatment with tebuconazole @ 1 g kg^{-1} and with commercial formulation of *Trichoderma harzianum* @ 5 g kg^{-1} seed along with soil application of neem cake @ 1.3 t ha^{-1} maintained its superiority over other treatments by recording the least PDI, maximum germination percentage (98.20%), root length (14.62 cm), shoot length (35.54 cm), number of pods per plant (32.57) and pod yield (3920.0 kg ha^{-1}). *P.* and *Bacillus* species have been reported effective against *M. phaseolina*. The mechanisms and antifungal compounds produced by these bacteria to control charcoal rot have been studied extensively (24, 25). This is particularly true if applied antagonists are able to proliferate sufficiently to colonize the germinating seeds and the developing roots. Besides, for successful root/rhizosphere colonization and plant growth promotion, *P. putida* should be able to tolerate the environmental conditions.

Conclusion

The PGPR *P. putida* (GN1) as seed treatment and soil application along with neem cake showed disease reduction of root rot and stem rot disease under field conditions for groundnut crop and also showed alteration of root architecture by increasing root length utilized for water absorption, root tips for protecting the root hairs and caps for drought and root diameter and root volume for drought survival. Therefore, it is recommended for plant growth promotion in different soil types on various crops.

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Authors' contributions

MP conceptualized the idea and obtained the project from Tamil Nadu Agricultural University, initiated the lab studies and conducted most of the experiments. AKB assisted in the field experiments. LR did formal analysis, MP and MK-Rhizoscanner study, AT statistical analysis, PI methodology, RR and IJ sequence analysis and JS review of the project.

Compliance with ethical standards

Competing Interests: All authors declared no conflict of interest

Ethical issues: None

References

1. FAOSTAT. Food and agricultural organization statistics database. [Internet]; 2019:7. Available from: https://www.fao.org/faostat/en/#home
2. INDIASTAT. Socio-economic statistical information about India. [Internet]; 2019:28. Available from: https://www.INDIASTAT.com
3. Choudhary A, Ashraf S. Utilizing the combined antifungal potential of *Trichoderma* spp. and organic amendments against dry root rot of mungbean. Egypt J Biol Pest Control. 2019;29(83). https://doi.org/10.1186/s41938-019-0187-8
4. Meena CM, Ashok KM, Prabhu NM, Raja RM. Management of stem rot of groundnut incited by *S. Rolfsii* through important bioagents. Chem Sci Rev Lett. 2018;7(28):1012-17.
5. Kumar V, Kumar A, Kharwar RN. Antagonistic potential of fluorescent pseudomonads and control of charcoal rot of chickpea caused by *Macrophomina phaseolina*. J Environ Biol. 2007;28(1):15-20. PMID: 17717979.
6. Paramasivan M, Kannan P, Rajendran L, Muthuramu S, Mytrle G. Management of root rot (*Macrophomina phaseolina*) in peanut with biocontrol agents and studying its root physiology. Arch Phytopathol Plant Prot. 2022;55(10):1169-78. https://doi.org/10.1080/03235408.2021.19688286
7. Zaman NR, Kumar B, Nasrin Z, Islam MR, Maiti TK, Khan H. Proteome analyses reveal *Macrophomina phaseolina's* survival tools when challenged by *Burkholderia contaminans* NZ. ACS Omega. 2020;5(3):1352-62. https://doi.org/10.1021/acsomega.9b01870
8. Dhawan A, Kumar S, Sharma PK, Chugh RK. Effect of different fungicides, organics amendments and bio-control agents on dry root rot of cluster bean (*Cyamopsis tetragonoloba* (L.) Taub) caused by *Rhizoctonia bataticola* (Taub.) Butler. Forage Res. 2019;44(4):276-81.
9. Vijayasanthi S, Akila R, Kannan R, Gomathy M. Evaluation of fungicides and oil cakes for the management of Panama wilt caused by *Fusarium oxysporum* f. sp. *cubense* (FOC) in banana. J Pharmacogn Phytochem. 2019;8(6):1258-63.
10. Rakholiya KB, Jadeja KB. Effect of soil amendments and bio-control agents for management of stem and pod rot of groundnut. Int J Plant Prot. 2010:3(2):348-49. UPENG/2008/24354.
11. Tayyab M, Islam W, Lee CG, Pang Z, Khalil F, Lin S. Short-term effects of different organic amendments on soil fungal composite. Sustainability. 2019;11(1):198. https://doi.org/10.3390/su11010198
12. Archana T, Rajendran L, Manoranjitham SK, Santhana Krishnan VP, Paramasivan M, Karthikeyan G. Culture-dependent analysis of seed bacterial endophyte, *Pseudomonas* spp. EGN 1 against the stem rot disease (*Sclerotium rolfsii* Sacc.) in groundnut. Egypt J Biol Pest Control. 2020;30:119. https://doi.org/10.1186/s41938-020-00317-x
13. Bonanomi G, Lorito M, Vinale F, Woo SL. Organic amendments, beneficial microbes and soil microbiota: Towards a unified framework for disease suppression. Annu Rev Phytopathol. 2018;56:1-20. https://doi.org/10.1146/annurev-phyto-080615-100046
14. Jangir M, Sharma S, Satyawati S. Synergistic effect of oilseed cake and biocontrol agent in the suppression of *Fusarium* wilt in *Solanum lycopersicum*. Braz J Microbiol. 2020;51:1929-39. https://doi.org/10.1007/s42770-020-00344-8
15. Vasebi Y, Safaie N, Alizadeh A. Biological control of soybean charcoal root rot disease using bacterial and fungal antagonists *In vitro* and greenhouse condition. J Crop Prot. 2013;2(2):139-50. http://jcp.modares.ac.ir/article-3-9001-en.html
16. Costa-Gutierrez SB, Adler C, Espinosa-Urgel M. *Pseudomonas putida* and its close relatives: mixing and mastering the perfect tune for plants. Appl Microbiol Biotechnol. 2022;106:3351–67. https://doi.org/10.1007/s00253-022-11881-7
17. Planchamp C, Glauser G, Mauch-Mani B. Root inoculation with *Pseudomonas putida* KT2440 induces transcriptional and metabolic changes and systemic resistance in maize plants. Front Plant Sci. 2015;5:719. https://doi.org/10.3389/ fpls.2014.00719
18. Saritha B, Panneerselvam P, Ganeshamurthy AN. Antagonistic potential of mycorrhiza associated *Pseudomonas putida* against soil borne fungal pathogens. Plant Arch. 2015;15:763–68.
19. Lurthy T, Cantat C, Jeudy C, Declerck P, Gallardo K, Barraud C, et al. Impact of bacterial siderophores on iron status and ionome in pea. Front Plant Sci. 2020;11:730. https://doi.org/10.3389/fpls.2020.00730
20. Daura-Pich O, Hernández I, Pinyol-Escala L, Lara JM, Martínez-Servat S, Fernández C, López-García B. No antibiotic and toxic metabolites produced by the biocontrol agent *Pseudomonas putida* strain B2017. FEMS Microbiol Lett. 2020;367(9). https://doi.org/10.1093/femsle/fnaa075
21. Oliver C, Hernández I, Caminal M, Lara JM, Fernàndez C. *Pseudomonas putida* strain B2017 produced as technical grade active ingredient controls fungal and bacterial crop diseases. Biocontrol Sci Technol. 2019;29:1053-68. https://doi.org/10.1080/09583157.2019.1645304
22. Wang R, Wang HL, Tang RP, Sun MY, Chen TM, Duan XC, et al. *Pseudomonas putida* represses JA- and SA-mediated defense pathways in rice and promotes an alternative defense mechanism possibly through ABA signaling. Plants. 2020;9(12):1641. https://doi.org/10.3390/plants9121641
23. Patricia B, Luke PA, Alain F, María AL. The *Pseudomonas putida* T6SS is a plant warden against phytopathogens. ISME J. 2017;11(4):972-87. https://doi.org/10.1038/ismej.2016.169
24. Castaldi S, Masi M, Sautua F, Cimmino A, Isticato R, Carmona M. *Pseudomonas fluorescens* showing antifungal activity against *Macrophomina phaseolina,* a severe pathogenic fungus of soybean, produces phenazine as the main active metabolite. Biomolecules. 2021;11(11):1728. https://doi.org/10.3390/biom11111728
25. Babu GN, Deepika DS. Integrated management of stem rot of groundnut caused by *Sclerotiumrolfsii*. sacc. Indian J Agric Res. 2023;57(2):235-41. https://doi.org/10.18805/IJARe.A-6043