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Mass multiplication of native rhizospheric bacteria on cheaply available organic substrates and their plant growth promotion potentiality on cowpea

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ABSTRACT

The intensive agricultural system results into considerable environmental damage and its inevitable effects on various environmental and ecological factors. Known for their role in the natural disease-suppressiveness of particular soil, *Fluorescent pseudomonads* are a genus of root-associated bacteria that may colonise the roots of agricultural plants and create antifungal metabolites, making them a viable alternative to the use of chemical fungicides. The successful application of *Fluorescent pseudomonads* as bioinoculants depends to a great extent on their capability to establish roots and to contend with indigenous microbiome in the rhizosphere. Because of various limitations, variations in the bio control agents are bound to occur in different agro-climatic regions, so it is advisable to choose native bio control agents. In the present investigation, a comprehensive effort was undertaken to check the growth of *Pseudomonas aeruginosa* (GP-8), which was found to be much lesser in aqueous extracts of cow dung, vermicompost and groundnut cake as compared to the growth of GP-8 (*Pseudomonas aeruginosa*) in nutrient broth media. The enrichment of Nitrogen and other supplementary nutrients may be required for the development of low cost indigenous media from cheaply available substrates for commercial mass production of *Pseudomonas aeruginosa* (GP-8) and also a field level screening trial was undertaken with efficient native *Fluorescent pseudomonads* solely or in consortia for improvement of plant health and management of soil borne pathogens as well as to develop a culture media with cheap and readily available substrate sources for mass production of *Fluorescent pseudomonads* strains. In the present study, it was found that shoot length and root length of native *Fluorescent pseudomonads* inoculated cowpea plants were significantly increased as compared with those of the control plants both at 30 and 60 days after sowing.

Key word: *Fluorescent pseudomonad, Microbial consortia, Collar rot disease of cowpea, growth promotion, Mass production*

Introduction

As the Green Revolution has revolutionized agriculture around the world, the scenario of crop production has changed significantly over time. The agriculture sector has witnessed major transformational

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changes from the earlier subsistence farming to the present day intensive agriculture system. In developing countries especially for the poor, the major protein sources in the diet are vegetable legumes. Because of their nutrient-rich mature pods and fodders makes cowpea one of the most significant leguminous vegetable crops. But nowadays cowpea production is hampered due to numerous biotic and abiotic stress factors. The average productivity of cowpea is comparatively low in West Bengal. The crops are infected by various diseases viz. collar rot, powdery mildew, rust, wilt, charcoal rot, stem rot, bacterial blight and different viral diseases. Aveling and Adandonon (2000) reported that one of the most important soilborne fungal pathogen of cowpea is *Sclerotium rolfsii,* responsible for collar rot disease. This disease results into significant losses in quality and yield. Different disease management strategies employed to manage this necrotrophic pathogen are application of chemical fungicides, soil solarization, deep ploughing, use of biocontrol agents, crop rotation and application of organic-inorganic componds (Punja, 1988).

The massive use of various chemical pollutants like improper use of heavy synthetic fertilizers and chemical pesticides caused havoc ecological imbalance and environmental effect with possible risks to human health. Continuous and non-judicious application of chemical fungicides poses significant side effects such as environmental contamination (Ongley, 1996), disease resistance development (Sparks, 2013; Tupe *et al*., 2014), residual toxicity issues (Yoom *et al*., 2013), and biodiversity losses (Ghorbani *et al*., 2008). Thus the intensification of farming system has a considerable effect on environment, soil eco-system and on soil health. The search for alternative, eco-friendly and economic methods for plant disease management is of paramount importance in our conditions and urgent necessity too.

The most promising of these possible soil microorganisms are the bacteria, popularly known as plant growth promoting rhizobacteria (PGPR). Among bacteria and fungi, several strains of rhizospheric microbial taxa such as *Bacillus* (Kumar *et al.,* 2015; Figueredo *et al.,* 2016), Pseudomonads (Vaja *et al.,* 2016; Kotasthane *et al.* 2017), *Streptomyces* (Li *et al.,* 2017), *Trichoderma*. (Rao *et al.,* 2015; Dania *et al.,* 2016) have proved as excellent biocontrol agents (BCAs) to manage various soil borne diseases.

Pseudomonas sp increases its attention both as BCAs and PGPRs due to its high competitive saprophytic ability, excellent root colonizing ability, metabolic diversity coupled with generation of plant growth regulators, siderophores, cyanogenic compounds and production of arrays of anti-fungal and anti-bacterial secondary metabolites including 2,4-diacetyl phloroglucinoal (DAPG), phenazines, pyrrolnitrin, pyoluteorin lipopeptides etc. (Raaijmaker *et al.,* 2002; Raaijmaker *et al.,* 2010).

The creation of sufficient amounts of PGPR strain is necessary for the development of successful formulation, so choosing an appropriate, affordable, and readily accessible medium is crucial. Because of the specific environmental requirements, it is frequently difficult to produce bacterial strains. Because of these limitations, variations in the biocontrol agents are bound to occur in different agro-climatic regions. Hence use of location- specific inoculation strains needs to be emphasized so as to obtain an optimum symbiotic benefit. Thus the selected medium for mass multiplication should be affordable, easily accessible, and have the right balance of nutrients.

Thus the present finding aims to evaluate different media developed from cheap and readily available organic sources for growth and mass production of efficient native rhizospheric fluorescent pseudomonads and the use of different native rhizospheric fluorescent pseudomonads and their microbial consortia with *Bacillus* sp. and *Trichoderma* sp. for improvement of crop health and management of collar rot disease of cowpea.

Materials and Methods

Native plant growth promoting rhizobacteria

Isolate of Fluorescent Pseudomonad (GP8: *Pseudomonas aeruginosa*) was collected from the Plant Bacteriological Laboratory, Department of Plant Pathology, BCKV and these strains were maintained by frequent sub-culturing using nutrient agar after 30 days interval and stored at 4^{o}C and used for different set of experiments.

Preparation of media with aqueous extract of organic resources

Various cheaply available organic resources like cowdung, vermicompost and groundnut cake were used at four different concentrations (5%, 10%, 15%, 20%) for preparation of the aqueous media for mass multiplication of fluorescent pseudomonads. 50 gm, 100 gm, 150 gm and 200 gm of respective organic resources were added to 1litre of distilled water to prepare the respective concentration. The flask were placed in orbital incubator shaker for 4hours and then allowed to settle down for next 1hour. The supernatant was filtered using membrane filter paper of size 0.4µ (Reddy *et al.*, 2012). The media were then kept for sterilization in the autoclave at 121.6 °C 15 psi for 20 min.

Determination of population of *Fluorescent pseudomonads* in various media composition containing cheap and readily available resources

Aqueous extracts of various organic substrates were mainly used at different concentration (5%, 10%, 15% and 20%). The pH was adjusted to 7.0 and a CFU (colony forming unit) of 1×10^5 cells/ml of GP-8 (*P. aeruginosa*), inoculated in 100 ml of all the rudimentarily designed media composition, and were incubated at 28±1 °C under agitation (150 rpm). The population of *P. aeruginosa* GP-8 was checked in samples after 48 h, by measuring the OD value at A600 and counting the viable CFU on King's B agar media. The viable count of the fluorescent pseudomonads on media was performed by serial dilution method and serial dilutions of the culture media after 10^{-7} was plated on petri plates containing specific medium and incubated in the BOD incubator at 28±1 °C for 48 h, after that the visible bacterial colonies appeared on the petri plates were counted.

Preparation of biocontrol formulation

Biocontrol formulation was prepared with talcum powder by following the procedure as described by Mallesh *et al.* (2018) was used. Bacterial suspension @25ml for each three bacterial isolate (GP8: *Pseudomonas aeruginosa*, S21SP14 : *Pseudomonas putida*, K11SP4 : *Pseudomonas baetica*), were mixed thoroughly with 250 g autoclaved talc powder along with 2.25 g carboxymethyl celulose. After drying the formulation overnight under aseptic condition, the talcum powder formulation was weighed and stored at room temperature. This formulation is then used as seed treatment and also for soil application.

Effect ofdifferent native rhizobacteria and their application on cowpea crop health under field condition

A field trial was conducted to test the potentiality of three native fluorescent pseudomonad isolates(GP8: *Pseudomonas aeruginosa*, S21SP14 : *Pseudomonas putida*, K11SP4 : *Pseudomonas baetica*), solely or in consortial mode (GP-8 + B11: *Bacillus subtillis*) (GP-8 + SAG 17A: *Trichoderma asperellum*) through seed as well as soil treatment at C-Block Research Farm ($22^0$5.7′ N latitude and $88^0$2.9′ E longitude with an elevation of 9.75 meters) of Bidhan Chandra Krishi Viswavidyalaya, for testing the performance of native rhizobacteria on growth promotion, yield and disease suppression of cowpea under natural conditions. Standard protocols were followed from sowing to harvesting of the crop. For field evaluation, seven treatments were taken with three replications including control; having individual plot size of 12 m^2. At the time of field preparation, FYM@2-3t/ha was applied followed by application of NPK @ 20:60:40 as basal dose. The periodical observation were recorded for various plant physical parameters like plant height, root length, number of branches, number of leaves, number of nodules, per plant. The data were recorded after 30 and 60 days after sowing and nodulation data was collected at 50 days after sowing and were analysed.

Results and Discussion

Mass multiplication of fluorescent pseudomonads in aqueous extracts of different locally available substrates

Aqueous extract of locally available substrate sources namely, vermicompost, cowdung manure and groundnut cake were used at four different doses (5%, 10%, 15% and 20%), where nutrient broth medium was kept as control. Viable cell count (CFU/ml) of GP-8 (*P. aeruginosa)* was recorded after 72 h of incubation at 28±1 ^{0}C.

The development of suitable formulations that enable the PGPR to remain viable for longer periods of time is a crucial component of biological control. Using liquid fermentation technique, King's B broth or nutrient broth is utilised to produce *Pseudomonas* and *Bacillus* species in large quantities (Manjula and Podile, 2001; Nakkeeran *et al.*, 2005). The data presented in the Table 2 indicated that irrespective of doses, the average growth of GP-8 (*Pseudomonas aeruginosa*) was only 59.36%, 27.81% and 18.62% respectively at aqueous extract of Groundnut Cake, vermicompost and cowdung as compared to the growth of GP-8 (*Pseudomonas aeruginosa)* in NB media (Table 1).

Based upon the organic waste material, vermi compost contains a variety of aliphatic and aromatic carbon, carbohydrate components, mono-saccharides, humic acid, fulvic acid, certain complex polysaccharides, and various nitrogenous organics in varying amounts (Roy, 2000; Canellas *et al*., 2010). These slowly metabolising saccharides are crucial for the long-term maintenance of the population of bacterial strain throughout its late log phase. According to Duffy and Defago (1999), the generation of antifungal metabolites by *Pseudomonas* has been significantly influenced by carbon, nitrogen, and mineral sources. This suggests that nutrient additions to formulations would be a good technique for increasing biocontrol efficacy. In cowdung media, composed of aqueous extract of cowdung, after incubation period, the growth of *P. aeruginosa* GP-8 was observed to be lowest as compared to the other substrates. In cowdung media, among the different doses the logarithmic growth of *P. aeruginosa* GP-8 was observed to be log CFU 9.33 /ml at 20% concentration which was even much lesser than the growth in NB medium (Table 1). Perusal of the data presented in tables indicated that the growth of *P. aeruginosa* GP-8 was in increasing trend with the increase in concentration of the locally available substrates. The significantly highest growth of *P. aeruginosa* GP-8 was observed in groundnut cake substrates followed by vermicompost and cowdung extract (Table 2). However, the growth of *P. aeruginosa* GP-8 was found to be much lesser in aqueous extracts of cowdung, vermicompost and groundnut cake and thus indicated that enrichment of nitrogen may be an important factors associated with the growth of GP-8 (*Pseudomonas aeruginosa*).

Table 1. Logarithmic population of GP-8 at different concentration (20%, 15%, 10%, 5%) and at different media (vermicompost, groundnut cake and cowdung)

Concentration	Logarithmic population of GP-8 (*Pseudomonas aeruginosa*)		
	Vermicompost cake	Groundnut cake	Cowdung cake
20%	9.47	9.75	9.33
15%	9.42	9.74	9.25
10%	9.25	9.63	9.07
5%	8.99	9.40	8.73
NB (Check)	9.87	9.88	9.88
SEM	0.015	0.018	0.057
CD*	0.048	0.056	0.178

Cost estimation of *Fluorescent pseudomonad* in aqueous extracts of different locally available substrates in comparison to Nutrient broth media

The data presented in Table 2, indicates that the percent growth of GP-8 is highest at 20% concentration in case of aqueous extract of all organic amendments. The cost incurred for preparing aqueous extract of various organic amendments viz., cowdung, vermicompost and groundnut cake is very minimum (Table 3), as compared to preparation for nutrient broth. For preparing 10l of media, the total ingredient cost incurred is Rs 12, Rs 24 and Rs 120 respectively (Table 3). But for preparing 10l of nutrient broth approximate cost of Rs 407.24 is required which is far more expensive as compared to the cheap readily available media. Hence it can be concluded that the aqueous extracts of groundnut with addition of some Nitrogen, Carbon and other mineral sources may be an effective economically cheap alternative media for large scale multiplication of fluorescent pseudomonads.

Effect of various plant growth promoting rhizobacteria (*Fluorescent pseudomonads*) and microbial consortia on shoot length, root length, number of branches and leaves and nodulation of cowpea plant

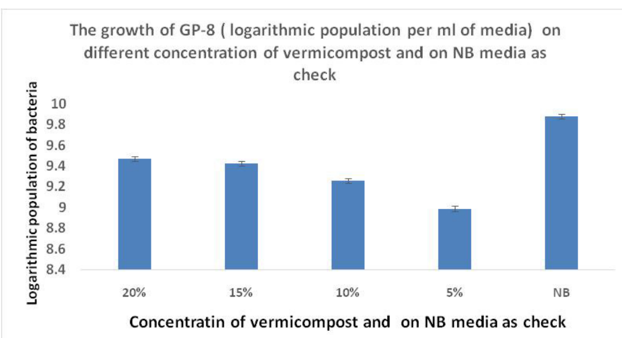
A field trial was conducted at C-Block Farm, BCKV, Kalyani, Nadia, West Bengal ($22^0 5.7'$ N latitude and

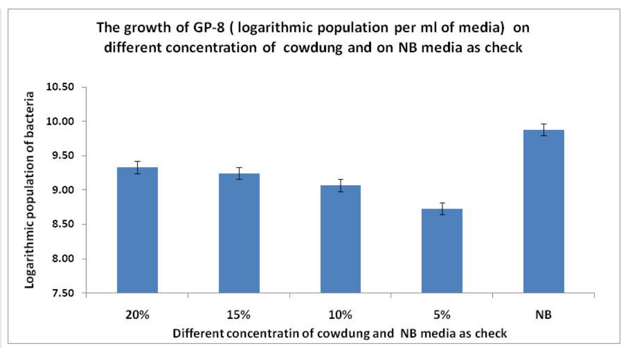
Table 2. Percent of GP-8 (*Pseudomonas aeruginosa*) at different concentration and at different media

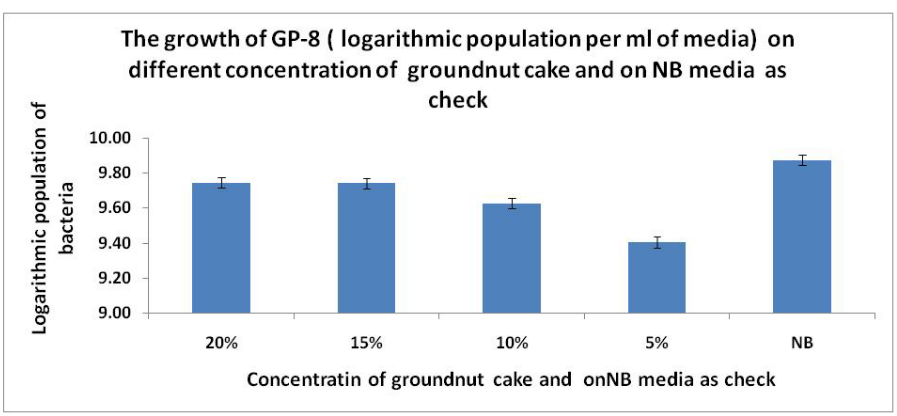
Name of the Organic Amendment	Percent growth of GP-8 (*Pseudomonas aeruginosa*) on different media prepared with organic amendment as compared to the growth on NB media — Concentration of Different Organic Amendment				
	20%	15%	10%	5%	Average
Vermicompost	38.98 (38.63)	35.27 (36.43)	23.95 (29.30)	13.04 (21.17)	27.81 (31.83)b
Groundnut Cake	73.95 (59.31)	73.39 (58.95)	56.09 (48.50)	34.02 (35.68)	59.36 (50.39)a
Cowdung	28.64 (32.36)	22.92 (28.60)	16.00 (23.58)	6.93 (15.26)	18.62 (25.56)c
Average	47.19 (43.39)a	43.86 (41.47)a	32.02 (34.46)b	17.99 (25.10)c	

88^{0}2.9′ E longitude) with cowpea (*Vigna unguiculata*), var. Kashi kanchan using seed treatment and soil application of bacterial isolates of fluorescent pseudomonads alone in three treatments (T1, T2, T3) and two treatments in combination, treatment T4, *Pseudomonas aeruginosa* + *Bacillus subtilis* (GP-8+B11) and treatment T5, *Pseudomonas aeruginosa* + *Trichoderma asperellum* (GP-8+SAG-17A). The influence of PGPR alone or in combination on cowpea were evaluated based on the seedling height, root length, no. of branches and leaves

Shoot length of the cowpea plants after 30 and 60 days after sowing was found maximum in the treatments T4 (25.53 cm, 37.73 cm), followed by T5 (23.47 cm, 35.20 cm) (Table 4) which comprised the consortial application of *Fluorescent pseudomonads* with *B.subtilis* (GP-8 + B11) and fluorescent pseudomonads with *T.asperellum* (GP-8 + SAG-17A), respectively through seed treatment as well as soil application. It may be concluded that the consortial application of fluorescent pseudomonads with *Bacillus subtilis* (GP-8+ B11) or with *Trichoderma asperellum* (GP-8 + SAG-17A) had synergistic effect on stimulation of the length of the cowpea plant. Among the three fluorescent pseudomonads used in the present study, two fluorescent pseudomonads namely S21SP14 and the GP-8 to be positive in ACC (1-aminocyclopropane-1-carboxylate) deaminase activity (Kumar, 2021). In the field experiment two fluorescent pseudomonads, namely S21SP14, GP-8 and consortia of fluorescent pseudomonads and *Bacillus subtilis* produced noticeably taller plants, indicating that increase in length is mainly due to ACC deaminase activity, lowering the ethylene levels in the plants and maximizing plant growth by ACC deaminase producing strains S21SP14, GP-8, consortia of fluorescent pseudomonads, and *Bacillus subtilis* (Glick *et al*., 1997; Penrose and Glick, 2001). This mechanism of plant growth promotion often has an impact on the growth of root hair, leading to structurally better rooting systems. Additionally, there is proof that the bacteria S21SP14 (*P. putida*), GP-8 (*P. aeruginosa*) of fluorescent pseudomonads, and *Bacillus subtilis* (B-11) are capable of producing the phytohormone IAA (Indole acetic acid), which may have contributed to the effects on growth promotion seen in the field experiments (Kumar, 2021). According to Yadav *et al.* (2010), *Pseudomonas aeruginosa* was the most effective inoculant for increasing root and shoot length and dry matter, fol-







Graph showing the growth of GP-8 (logarithmic population per ml of media) on different concentration (5%, 10%, 15%, 20%) in different media (vermicompost, cowdung, groundnut cakes) and on Nutrient Broth media as check lowed by *Bacillus subtilis*, *B. polymyxa*, *Paenibacillus* and *Bacillus boronophillus* over control. The present findings were consistent with their findings.

Bacterial inoculation significantly enhances ($P < 0.05$) the root length (cm) of the cowpea plants compared to the control both after 30 and 60 days after sowing. Average root length enhancement was 31.1, 25.7, 22.3 and 16.2 % compared to control at 30 days after sowing when (GP-8+ B11), GP-8, (GP-8 + SAG-17A), and S21SP14 were applied, respectively, through seed treatment as well as soil application. The average root length (cm) of the cowpea plants after 60 days of sowing was found to be maximum in the treatments T1 (19.17 cm), followed by T4 (18.43 cm) (Table 4). The present finding was in relation with the findings of Kumar *et al*. (2007) who observed that some potential isolates of fluorescent pseudomonads also significantly increased shoot length, root length of chickpea seeds. Jarak *et al*. (2012) also reported that co -inoculation of multiple rhizobacteria significantly improved plant health, as evidenced by increases in seedling emergence and vigour. Similar research was performed to examine seed bacterization with the isolate *Pseudomonas spp*. which increased the root growth more than the shoot growth, in comparison to control (Mishra *et al*., 2009).

Inoculation with fluorescent pseudomonads solely or in combination with *T. asperellum* (SAG-17A) or *B. subtilis* (B11) significantly enhances ($P < 0.05$) the number of branches and leaves of the cowpea plants compared to the control both after 30 and 60 days after sowing. It was also observed that the maximum number of leaves of the cowpea plants was observed in T4 (21.47 and 29.73) followed by T5 (21.13 and 28.18), respectively on 30 and 60 days after sowing. The percent increase of no. of leaves over inoculated plants was calculated and found as 51, 43.3, 31 and 27.6 % compared to the control at 60 days after sowing when (GP-8+ B11), (GP-8 + SAG-17A), GP-8 and S21SP14 were applied, respectively, through seed treatment as well as soil application. Consortial application of *P. aeruginosa* with *B. subtilis* (GP-8+B11) induces the maximum number of nodule per plant (46.33), followed by GP-8 (42.33).

Thus it may be concluded that the consortial application of fluorescent pseudomonads with *B.*

Table 3. Comparison of the production cost of *Fluorescent pseudomonads* biomass in respect to Nutrient Broth composition media and different cheaply available subsrate media

Composition	Nutrient broth		Different cheaply available substrate		
	Quantity required per 10l	Cost per 10l(Rs)	Composition per 10l	Quantity required	Cost per 10l(Rs)
Peptone	50gm	299.7	Vermicompost	2kg	24
Yeast extract	15gm	42.54	Groundnut cake	2kg	120
Beef extract	15gm	45	Cowdung	2kg	12
Sodium chloride	50gm	20			
Total	407.24				

Table 4. Effect of various plant growth promoting rhizobacteria (*Fluorescent pseudomonads*) and microbial consortia on shoot length, root length, number of branches and leaves and nodulation of cowpea plant

Treatments	Shoot length		Root length		No. of branch/pl.		No. of leaves/pl.		No. of nodule/pl
	30 DAS	60 DAS	30 DAS	60 DAS	30 DAS	60 DAS	30 DAS	60 DAS	50 DAS
T1: GP-8	21.40	32.63	8.40	19.17	4.67	6.50	18.67	25.77	42.33
T2: S21SP14	20.73	31.77	7.76	17.87	4.83	6.33	18.87	25.10	40.00
T3: K11SP4	18.93	28.87	7.45	15.87	4.33	5.67	17.33	22.67	35.33
T4: GP-8 + B11	25.53	37.73	8.76	18.43	5.50	7.50	21.47	29.73	46.33
T5: GP-8 +SAG-17A	23.47	35.20	8.17	18.20	5.33	7.17	21.13	28.18	40.33
T6: Carbendazim 50%WP @1g/kg	17.80	26.43	7.08	15.23	4.33	5.33	16.90	21.17	27.67
T7: Control	16.73	24.80	6.68	14.67	4.17	5.00	16.27	19.67	27.00
SEM	1.005	1.65	0.27	0.49	0.25	0.300	0.96	1.12	1.02
CD* (P=0.05)	3.095	5.09	0.84	1.51	0.78	0.924	2.94	3.46	3.14

subtilis or with *T.asperellum* had synergistic effect on stimulation of the shoot length, root length, number of branches and leaves, nodules per plant. Increases in fresh plant weight by the utilization of native PGPRs were also recorded by Prashant *et al*. (2009) in wheat plants. Zahir *et al*. (2008) also reported that inoculation with *Pseudomonas fluorescens* increased in root and shoot length, number of leaves per plant of pea plants and fresh and dry weight. Similar findings were also recorded by Samy *et al*. (2007) who reported that inoculation with *Pseudomonas aeruginosa* increased the number of pods per plant, shoot dry weight and nodule dry weight of faba bean.

Effect of various plant growth promoting rhizobacteria (*Fluorescent pseudomonads*) and microbial consortia on yield of cowpea plant

Application of the native *Fluorescent pseudomonads* (GP8, S21SP14, and K11SP4) alone or in combination with *Trichoderma asperellum* (SAG-17A) or *Bacillus subtilis* (B11) significantly increases ($P \leq 0.05$) the pod yield as compared with the uninoculated control. The relative increase in yield characteristics with regards to control ranged from 18.32% to 41.31% for pod yield. It was observed that the highest yield of cowpea was observed in T4 (11.80 t/ ha) followed by T5 (11.28 t/ ha) (Table 5). It may be concluded that the consortial application of fluorescent pseudomonads with *B. subtilis* or with *T.asperellum* had synergistic effect on the increases in yield of cowpea. The results of Di Salvo *et al*. (2018), who reported that PGPR applied as inoculants over cereal crops, like maize, can boost their growth and grain yield, were in agreement with the findings of the current study. According to Mishra *et al*. (2014), *Pseudomonas putida* PGRs improved the French bean's root and shoot dry weight, nodulation, nutrient uptake, and pod yield.

Table 5. Effect of various plant growth promoting rhizobacteria (*Fluorescent pseudomonads*) and microbial consortia on yield of cowpea

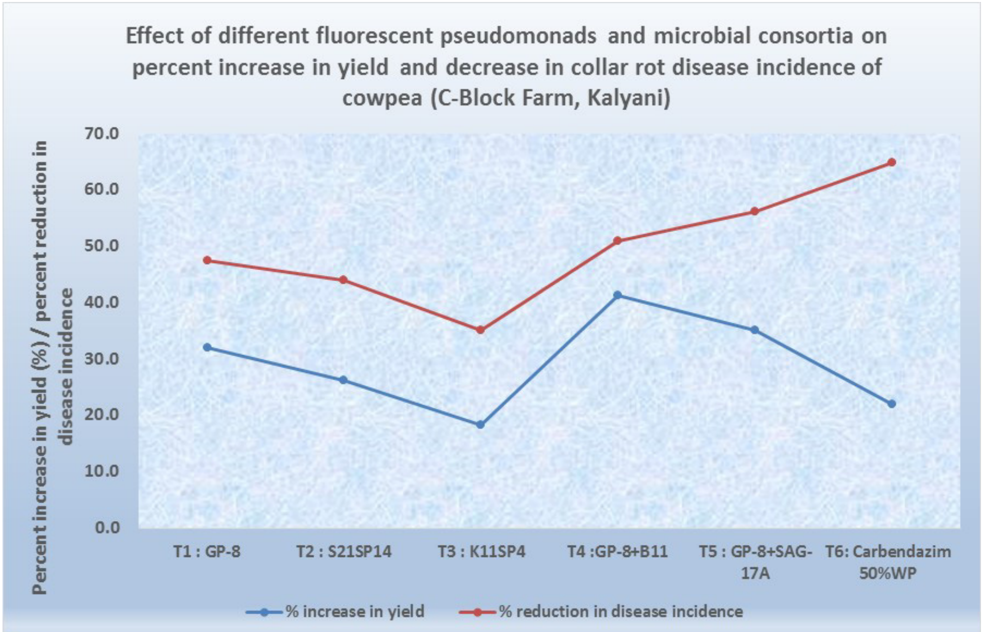
Treatments		Yield (t/ha)
T1:	GP-8	11.03
T2:	S21SP14	10.54
T3:	K11SP4	9.88
T4:	GP-8 + B11	11.80
T5:	GP-8 +SAG-17A	11.28
T6:	Carbendazim 50%WP @1g/kg	10.19
T7	Control	8.35
	SEM	0.257
	CD* (P=0.05)	0.793

Effect of various plant growth promoting rhizobacteria (*Fluorescent pseudomonads*) and microbial consortia on collar rot disease incidence of cowpea

Fluorescent Pseudomonads were found to suppress a number of soil borne diseases including *S.rolfsii* as reported by Mukhopadhyay et al. (1992) and proved to be effective biocontrol agents because by colonising the rhizosphere and promoting quick nutrient absorption, and enhance plant development. Fluorescent Pseudomonads has the biocontrol ability to fight against various soil- and root-borne diseases in variety of crops, including tomato, wheat and chickpea (Grover *et al*. 2009; Dashti *et al*. 2012; Perez-Montano *et al*. 2014). It was revealed from Table 6 that in the present study. The incidence of collar rot disease significantly reduces ($P<0.05$) in all treatments as compared to un-treated control. The least incidence of collar rot disease was observed in fungicide treated plot T6 (6.67 %) followed by T5 (8.33%). The highest percentage decrease in disease

Table 6. Effect of various plant growth promoting rhizobacteria (*Fluorescent pseudomonads*) and microbial consortia on collar rot disease incidence of cowpea

	Treatments	Disease incidence (%)	Percent disease control
T1:	GP-8	10.00 (18.43)	47.4
T2:	S21SP14	10.67 (19.06)	43.9
T3:	K11SP4	12.33(20.56)	35.1
T4:	GP-8 + B11	9.33(17.79)	50.9
T5:	GP-8 +SAG-17A	8.33(16.78)	56.1
T6:	Carbendazim 50%WP	6.67(14.96)	64.9
T7	Control	19.00(25.84)	
	SEM	0.74	
	CD* (p=0.05)	2.28	



incidence (DI%) over control was observed in T6 (64.9 %), followed by T4 with 56.1 %. However, no significant difference was observed in collar rot disease incidence in T6- fungicide treated plot and T5 (GP-8+ SAG17A) treated plots.

The increase in various growth parameters of PGPR treated plants in the current study may be due to seed biopriming as well as soil application with bacterial antagonists leads to an increase in their population with time in the surrounding of seed which protected them from attacks by different plant pathogens (Callan *et al.*, 1990).

Economics of application of Plant Growth Promoting Rhizobacteria

The average price of cowpea vegetable pod was Rs. 10 per kg. Economics of the application of PGPR was then calculated based on the yield data. The maximum incremental cost benefit ratio (1:10.4) was recorded under T4(GP-8+ B11) and which was followed by 1:8.7 at T_5 (GP-8+SAG-17A) (Table 7).

Conclusion

In the current agricultural systems, for the management of biotic and abiotic stresses without harming the soil and environment, there is an increasing demand for bio-rational techniques. The application of PGPR may be the key for improving soil and plant health under sustainable agricultural system. In the present investigation, a comprehensive effort was undertaken for field level screening of the efficient native *Fluorescent pseudomonads* solely or in consortia for improvement of plant health and manage-

Table 7. Economics of use of different native *Pseudomonas sp.* in the field trial:

	Treatment	Cost of application (Rs/ha)*	Yield (t/ha)	Additional yield over control (t/ha)	Additional profit from produce (Rs/ha)#	Net profit (Rs/ha)	Cost: Benefit Ratio (ICBR)
T1	GP-8	2882.00	11.03	2.68	26800.00	23918.00	1:8.3
T2	S21SP14	2882.00	10.54	2.19	21900.00	19018.00	1:6.6
T3	K11SP4	2882.00	9.88	1.53	15300.00	12418.00	1:4.3
T4	GP-8 + B11	3015.75	11.80	3.45	34500.00	31484.30	1:10.4
T5	GP-8 +SAG-17A	3015.75	11.28	2.93	29300.00	26284.30	1:8.7
T6	Carbendazim 50% WP	2140.00	10.19	1.84	18400.00	16260.00	1:7.6
T7	Control	-	8.35	-	-	-	-

ment of soil-borne pathogens in Gangetic alluvial regions and attempts were also taken to develop a media with cheap and readily available substrate sources for mass production of fluorescent Pseudomonads strains. The growth of *P. aeruginosa* GP-8 was found to be much lesser in aqueous extracts of cowdung, vermicompost and groundnut cakeas compared to the growth of GP-8 *(Pseudomonas aeruginosa)* in NB media. Irrespective of doses, the average growth of GP-8 was only 59.36%, 27.81% and 18.62% respectively at aqueous extract of groundnut cake, vermicompost and cowdung as compared to the growth of GP-8 in NB media. The findings, thus indicated that enrichment of Nitrogen and other supplementary nutrients may be added to the cheaply available substrates for mass production of *Pseudomonas aeruginosa* (GP-8). Two of the fluorescent pseudomonad isolates were shown to considerably improve cowpea plant growth features and yield when their use was evaluated for its potential in sustainable agriculture. The highest incremental cost benefit ratio in a crop cycle were also being recorded. However, from the experiment it may be concluded that the consortial application of fluorescent pseudomonads with *Bacillus* sp or with *Trichoderma* sp had synergistic effect on stimulation of the increases in yield characteristics of cowpea.

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Conflict of Interest

No conflicting interest.

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