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Host Specific Plant Growth Promoting Activity of IAA Producing and Phosphate Solubilizing Fluorescent *Pseudomonas*

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A B S T R A C T

Fluorescent *Pseudomonas* possesses many traits that make them well suited as biocontrol and growth promoting agents. Host specific plant growth promoting activity of fluorescent *Pseudomonas* isolates was observed. Isolates #P72, P141, P151, P233, P124, P6, P143, P176, P76, P99, P167 were able to induce the formation of increased root and shoot length. Isolates used in the present investigation had the ability (although in different proportions) to solubilize inorganic phosphate, produce Indole acetic acid (IAA) and PHB. Frequency of fluorescent *Pseudomonas* isolates which induced shoot length of crop plants more than fluorescent *Pseudomonas* isolates with the ability to induced root length. It was also observed that Fluorescent *Pseudomonas* isolates reduced root shoot length as compared un-treated control.

Introduction

Microorganisms, interact with higher plants in soil ecosystem and influences the development of plant root in the soil (Ahmad *et al.*, 2008; Taghavi *et al.*, 2009). Beneficial plant-microbe interactions enhance plant growth and nutrient uptake by different mechanisms, including increased mobilization of insoluble nutrients (Lifshitz *et al.*, 1987; Ahmad *et al.*, 2008), biocontrol of phytopathogenic organisms (Weller, 2007) and/or by production of phytohormones (Dubeikovsky *et al.*, 1993; Spaepen *et al.*, 2007).

Pseudomonas putida are ubiquitous bacteria frequently present in the plant rhizosphere

(Timmis 2002; Dos Santos *et al.,* 2004) and possess many traits that make them well suited as biocontrol and growth promoting agents (Fravel, 1988; Lemanceau, 1992; Weller *et al.,* 2002; Fravel, 2005) because of their potential to produce secondary metabolites (Leisinger and Margraff, 1979), growth hormones (Brown 1974), antibiotics (Fravel, 1988; Weller *et al.,* 2002) and chelating compounds (siderophores) (Leong, 1986). Phosphate solubilization is one of the direct mechanisms (Rodríguez and Fraga , 1999; Mayak *et al.,* 2004; Shahzad *et al.,* 2010) and production of antibiotics such as 2,4-diacetyl phloroglucinol (DAPG), phenazine, pyoluteorin and pyrrolnitrin against pathogenic fungi and bacteria are among indirect mechanisms of PGPR (McSpadden Gardener *et al.,* 2001; Ramamoorthy *et al.,* 2001). Some of them may also be involved in the biodegradation of natural or man-made toxic chemical compounds(Holloway 1992; Ramos *et al.,* 2009). *P. putida* show diverse spectrum of metabolic versatility and niche-specific adaptations (Rojo 2010; Wu *et al.,* 2011). Several lines of evidence suggest that PGPB produced IAA which may directly stimulates plant growth, even in the presence of otherwise inhibitory compounds (Wani *et al.,* 2008; Bianco and Defez 2009; Egamberdieva 2009; Bianco and Defez 2010; De-Bashan *et al.,* 2010). IAA synthesized by bacteria may be involved at different levels in plant-bacterial interactions. Fluorescent *Pseudomonas* may alter the suboptimal or optimal endogenous IAA level in plant roots(Pilet and Saugy 1987) to either optimal or supraoptimal, resulting in plant growth promotion or inhibition, respectively. IAA-deficient mutant of *Pseudomonas putida* GR12-2(Patten and Glick 2002a) had a reduced root length as compared to wild-type *P. putida* GR12-2 which induced the formation of roots that were 35–50% longer thus suggesting the role of bacterially produced IAA in root development. On the other hand IAA positive mutants (overproducing) of same strains (*Pseudomonas putida* GR12-2) (Xie *et al.,* 1996), yielded greater number of shorter roots on inoculation of mung bean cuttings as compared to wild type strain. Combined effect of auxin on growth promotion and inhibition of root elongation by ethylene has been reported (Jackson 1991). Apart from primary and secondary metabolite production, certain fluorescent Pseudomonads (especially *P. putida*) are suitable as whole-cell biocatalyzers for the production of several value-added industrial compounds such as biodegradable and biocompatible polyesters called polyhydroxyalkanoates (PHA) or polyhydroxybutyrates (PHB). It accumulates as discrete granules and is used as storage material for carbon and for reducing equivalents by *P. putida*. This property has been widely exploited for their targeted biosynthesis in this organism (Hoffmann and Rehm 2004). Different strains of *P. putida* such as *P. putida* KT2440, *P. putida* GPo1, *P. putida* S12, etc. have been investigated for its capacity to accumulate PHAs and PHBs from different carbon sources (Durner *et al.,* 2001; Hartmann *et al.,* 2004; Meijnen *et al.,* 2008). The pha gene cluster is responsible for the accumulation of PHAs and PHBs in *P. putida* (Vo *et al.,* 2008; Chung *et al.,* 2009; Wang and Nomura 2010). In the present investigation the potential of IAA producing and phosphate solubilizing fluorescent *Pseudomonas* on Bottlegourd, Chickpea, Greengram, Lathyrus, Rice, Wheat was studied.

Materials and Methods

Bacterial isolates

The experimental material consisted of twenty four isolates of Fluorescent *Pseudomonas spp* isolated from soil (rhizospheric and non-rhizospheric) samples of different

geographical locations of Chhattisgarh (Table 1). Isolated bacterial colonies after incubation at 28°C for 2 days, were exposed under UV light (366 nm), emitted fluoresces from the colonies and biochemical tests as per the procedures outlined in Bergey's Manual of Systematic Bacteriology (Sneath *et al.*, 1986) confirmed the identity as fluorescent *Pseudomonas*. Culture were maintained on King's B broth (Himedia) containing 50% (w/v) glycerol at -80°C in the Department of Plant Molecular Biology and Biotechnology, Indira Gandhi Krishi Vishwavidyalaya, Raipur, Chhattisgarh, and were revived on King's B slants as and when required.

Screening for polyhydroxybutyrate (PHB) production and its quantitative estimation

Fluorescent *Pseudomonas* isolates were screened for PHB accumulation qualitatively by following the viable colony method using Sudan Black B dye (Liu *et al.*, 1998). Sterilized Nutrient agar (Himedia) supplemented with 1 % glucose was spot inoculated with the isolates and incubated at 30°C for 24 h. Ethanolic solution (0.02 %) of Sudan Black B was spread over the colony and the plates were kept undisturbed for 30 min. Later, they were washed with ethanol (96 %) to remove the excess stain from the colony. The dark blue colored colony was taken as positive for PHB production. The Sudan Black B positive isolates were subjected to quantification of PHB production as per the method of (Law and Slepecky 1961). The bacterial cells containing the polymer were pelleted at 10,000 rpm for 10 min. and the pellet was washed with acetone and ethanol to remove the unwanted materials. The pellet was re-suspended in equal volume of 4 % sodium hypochlorite and incubated at room temperature for 30 min. The whole mixture was again centrifuged and the supernatant was discarded. The cell pellet containing PHB was again washed with acetone and ethanol. Finally, the polymer granules were dissolved in hot chloroform. The chloroform was filtered and to the filtrate, concentrated 10 ml hot H_2SO_4 was added. The addition of sulfuric acid converts the polymer into crotonic acid which is brown colored. The solution was cooled and the absorbance was read at 235 ηm against a sulfuric acid blank. By referring to the standard curve prepared using Poly[(R)-3-hydroxybutyric acid] (Sigma Aldrich, USA) by following the method of (Law and Slepecky, 1961), the quantity of PHB produced by different bacterial isolates was determined.

Determination of Indole Acetic acid (IAA) production and phosphate solubilization by *Pseudomonas* spp.

For the quantitative estimation of IAA, active culture of *Pseudomonas* spp. were inoculated to 20 ml DF salts minimal media (Dworkin and Foster 1958) in 100 ml conical flasks and incubated for 3 days at 28°C. The medium was supplemented with L-tryptophan at a concentration of 1.02 g/l. After incubation for 72 h, the grown bacterial cells were removed from the culture medium by centrifugation at 5,000 rpm for 5 min and the pH of the medium of all isolates was recorded. One ml aliquot of the supernatant was mixed vigorously with 4 ml of Salkowski's reagent (Gordon and Weber 1951) and allowed to stand at room temperature for 20 min. The absorbance at 535 ηm was measured with DF salts minimal media (plus Salkowski's reagent) as blank. The concentration of IAA in each culture supernatant was determined using an IAA (Himedia) standard curve.

Screening of phosphate solubilisation ability and its quantitative estimation

Qualitative screening of phosphate solubilising fluorescent *Pseudomonas* was performed on Pikovskaya agar medium

(Himedia) containing tricalcium phosphate as a phosphate source and bromocresol purple (0.1 g/l) as a pH indicator for acidification(Vazquez *et al.,* 2000). After incubation of fresh cultures of fluorescent *Pseudomonas* at 28 ± 2 _C for 48 h, phosphate solubilizing isolates turned the media color from purple to yellow in the zones of acidification.

Quantitative estimation of phosphate solubilisation in Pikovskaya broth (Himedia) was performed according to the procedure of (Murphy and Riley 1962). Fresh cultures of fluorescent *Pseudomonas* isolates were inoculated to 50 ml of Pikovskaya's broth and incubated at 28 ± 2 _C and 100 rpm. The amount of inorganic phosphate (Pi) released in the broth was estimated after 7 days of incubation in comparison with un-inoculated control.

The broth culture was centrifuged at 10,000 rpm for 10 min to separate the supernatant from the bacterial growth and insoluble phosphate. To the 0.5 ml of the culture supernatant 5 ml of chloromolybdic acid was added and mixed thoroughly.

Volume was made up to 10 ml with distilled water and 125 ll of chlorostannous acid was added to it. Immediately, the final volume was made up to 25 ml with distilled water and mixed thoroughly. After 15 min, the blue color developed was read in a spectrophotometer at 610 nm using a reagent blank.

Corresponding amount of soluble phosphorous was calculated from standard curve of potassium dihydrogen phosphate (KH2PO4). Phosphate solubilizing activity was expressed in terms of tricalcium phosphate solubilization which in turn was measured by lg/ml of available orthophosphate as calibrated from the standard curve of KH2PO4.

Results and Discussion

Quantification of Indole acetic acid (IAA) production by *Pseudomonas spp.*

Production of IAA and IAA related compounds was evaluated for twenty four isolates of fluorescent *Pseudomonas spp.* in DF salt culture medium amended with 1.02 g/L from 5mM stock of L-tryptophan as precursor molecule and any IAA precursor molecule as control. The mixture of culture supernatant and Salkowski's reagent was incubated at room temperature for 20 min and the absorbance was measured at 535 ηm. The concentration of IAA and IAA related compounds was evaluated by comparison with a standard curve prepared using serial dilutions (0-100 µg/ml) prepared from commercially available IAA. Interpolation of the colorimeter readings with standard curve were used to quantify the amount of IAA produced by different isolates of *Pseudomonas* in the media which ranged from 8.09 to 63.18 µg/ml (Table 1).

Twenty four fluorescent *Pseudomonas* isolates produced varying proportions of IAA from L-tryptophan as a precursor. Fluorescent *Pseudomonas* isolates P124, P99, P72and P201 secreated high proportions of IAA in the supernatents. Maximum amount of IAA production was observed with isolate P124 (63.18 µg/ml) whereas isolate P6 (8.09 µg/ml) was the lowest producer (Table 1). Three groups of IAA producers could be resolved after interpolation of the colorimeter readings with standard curve were used to quantify the amount of IAA secreted by different isolates of *Pseudomonas* in the media: Low IAA secreting fluorescent *Pseudomonas* isolates (8.09 to 9.91(µg/ml)) were P6 P233 P85 P167 and P216; Medium IAA secreting fluorescent *Pseudomonas* isolates (10.27 to 16.18(µg/ml)) were P141, P76, P176, P143, P247, P179, P11, P151, P126, P67, P248, P5, P161) and

High IAA secreting fluorescent *Pseudomonas* isolates (21 to 63 (µg/ml)) were P129, P205, P201, P72, P99, P124 (Table 1).

Several PGPRs as well as some pathogenic, symbiotic and free living rhizobacterial species are reported to produce IAA and gibberllic acid in the rhizospheric soil and thereby plays a significant role in increasing the root surface area and number of root tips in many plants (Han *et al.,* 2005). Recent investigations on auxin synthesizing rhizobacteria (Spaepen *et al.,* 2007) as phytohormone producer demonstrated that the rhizobacteria can synthesize IAA from tryptophan by different pathways, although the general mechanism of auxin synthesis was basically concentrated on the tryptophan-independent pathways. The phytopathogenic bacteria rather use the indole acetamide pathway to synthesize IAA that has been implicated earlier in the tumor induction in plants. (Swain *et al.,* 2007) reported a positive effect of IAA producing strains of Bacillus subtilis on Dioscorea rotundata L. They applied a suspension of *B. subtilis* on the surface of the plant, which resulted in an increase in the root: stem ratio as well as number of sprouts as compared with the non-inoculated plants. Potentiality of *Azotobacter* spp., to produce high amount of IAA (7.3–32.8 mg/ml) in agriculture was reported by (Ahmad *et al.,* 2005). Laboratory studies have emphasized on use of plant growth promoting rhizobacteria (PGPR) as biocontrol agents (Hossain *et al.,* 2007) and the role of auxin (IAA) in plant growth promotion (Contreras-Cornejo *et al.,* 2009). Fluorescent *Pseudomonas* are one of the most abundant bacteria in the rhizosphere of many plants (Freitas and Germida 1990; Botelho and Mendonça-Hagler 2006), have large capacity to produce phytohormones, mainly auxins (Patten and Glick 1996; Patten and Glick 2002a; Patten and Glick 2002b; Khalid *et al.,* 2005) and secondary metabolites, such as antibiotics (Bergsma-Vlami *et al.,* 2005), thus they are able to improve plant growth and plant health (Glick 1995; Belimov *et al.,* 2007; Belimov *et al.,* 2009a; Belimov *et al.,* 2009b).

Screening of phosphate solubilizing *Pseudomonas spp.* and its quantification

Phosphorus frequently is the least accessible macronutrient in many ecosystems and its low availability is often limiting to plant growth (Raghothama, 1999). All the 24 isolates were capable of differentially utilizing tri-calcium phosphate in both agar plate and broth assays. By preparing standard curve the amount of inorganic phosphate solubilized by different *Pseudomonas* isolates were estimated wavelength 610ηm. Screening of isolates showed variation in their ability to utilized calcium phosphate supplemented in different nutrient constituents. Qualitative and Quantitative estimation of phosphate solubilization, carried out after incubation of 7 days at 28±2°C is presented in (Table 1). *In vitro* phosphate solubilization efficacy of fluorescent *Pseudomonas spp.* isolates as performed on Pikovskaya's agar by acidification showed positive results for all the 24 isolates tested.

Quantitative estimation of soluble phosphate concentrations in Pikovskaya's broth was expressed as µg/ml and it varied significantly from 88 to 768µg/ml.

Isolate P216 can be considered as promising inducer of phosphate mobilization. The amount of inorganic phosphate solubilized was 768 µg/ml followed by isolates P67, P201 P72 P76 and P161solubilizing 550 µg/ml, 518 µg/ml and 502 µg/ml, 484 µg/ml, 455µg/ml phosphate. Among 24 isolates screened these were the best phosphate solubilizers (Table 1).

These candidate isolates can be used as microbial inoculants to improve soil fertility

by releasing bound phosphorus thereby increasing the crop yield potential. Stimulation of different crops by plant growth promoting *Pseudomonas* isolates with potential phosphate solubilization ability may help in exploiting large reserves of phosphorus present in most agricultural soils. Several *Pseudomonas* species have been reported among the most efficient phosphate-solublizing bacteria and as important bio-inoculants due to their multiple biofertilizing activities of improving soil nutrient status, secretion of plant growth regulators and suppression of soil-borne pathogens (Rodríguez and Fraga 1999; Gulati *et al.,* 2008; Agrawal *et al.,* 2015).

Screening of polyhydroxybutyrate (PHB) producers and its quantification

Biodegradable and biocompatible polyesters such as polyhydroxyalkanoates (PHA) have potential pharmaceutical values (Takahashi *et al.,* 1994). In an alkaline environment *Pseudomonas* has been reported to produce medium-chain-length (R)-3-hydroxy alkanoates (Wang *et al.,* 2007). In the present investigation all the 24 fluorescent *Pseudomonas spp.* isolates gave positive result for PHB accumulation in Sudan Black B qualitatively screening test in 1%glucose supplemented nutrient agar medium. (Madison and Huisman, 1999) have also reported that these biopolymers are accumulated as inclusions (PHA granules) in the bacterial cytoplasm in response to inorganic nutrient limitations, generally, when the microbes are cultured in the presence of an excess carbon source.

However, in present study there was significant difference in quantitative analysis of all the isolates which ranged from 2.67 to 15.74mg/ml. The lowest observed value was for isolate P6 (2.67 mg/ml) and the highest was for isolate P126 (15.74 mg/ml). Isolates P99, P161, P233, P151 and P179 also produced significantly higher amounts of PHB (i.e 15.31 mg/ml, 14.52 mg/ml, 14.40 mg/ml and 14.25 mg/ml and 14.05 mg/ml respectively) as compared to other isolates (Table 1).

Two catagories of PHB producers were resolved after 24 fluorescent *Pseudomonas spp.* isolates gave positive result for PHB accumulation in Sudan Black B qualitatively screening test in 1%glucose supplemented nutrient agar medium PHB production 2.67 to 8.99 (mg/ml) P6, P11, P5, P201, P67, P124, P85; PHB production 10.48 to 15.75 (mg/ml) P143, P141, P205, P216, P167, P247, P76, P72, P248, P129, P176, P179, P151, P233, P161, P99, P126

Apart from primary and secondary metabolite production, certain fluorescent Pseudomonads (especially *P. putida*) are suitable as whole-cell biocatalyzers for the production of several value-added industrial compounds such as biodegradable and biocompatible polyesters called polyhydroxyalkanoates (PHA) or polyhydroxybutyrates (PHB). It accumulates as discrete granules and is used as storage material for carbon and for reducing equivalents by *P. putida*. This property has been widely exploited for their targeted biosynthesis in this organism(Hoffmann and Rehm, 2004). Different strains of *P. putida* such as *P. putida* KT2440, *P. putida* GPo1, *P. putida* S12, etc. have been investigated for its capacity to accumulate PHAs and PHBs from different carbon sources (Durner *et al.,* 2001; Hartmann *et al.,* 2004; Meijnen *et al.,* 2008). The pha gene cluster is responsible for the accumulation of PHAs and PHBs in *P. putida* (Vo *et al.,* 2008; Chung *et al.,* 2009; Wang and Nomura 2010). Expenditures for large-scale production of PHA were almost evenly divided between carbon source, fermentation process and separation process (Sun *et al.,* 2007; Elbahloul and Steinbüchel 2009).

Therefore, screening for carbohydrate utilization by *Pseudomonas* isolate may help in identifying candidate isolate which dwells upon cheaper carbon sources. Our work reports that all the 24 fluorescent *Pseudomonas* isolates utilized relatively cheaper carbohydrates such as xylose, dextrose, galactose, melibiose, and mannose corroborates earlier reports by Agrawal *et al.*, (2015).

Plant growth promoting response of rice, wheat, greengram, blackgram, lathyrus, chickpea, bottle gourd following seed bacterization with fluorescent *Pseudomonas* isolates

Indole acetic acid affects the physiology of plants in dramatically different ways which is a well know fact. Plant responses to IAA vary from plant to plant tissue involved (roots, shoots, the optimal level of IAA for supporting plant growth is ~5 orders of magnitude lower for roots than for shoots); and as a function of the developmental stage of the plant. However, the endogenous pool of plant IAA may be suboptimal or optimal (Pilet and Saugy 1987) may be altered by the acquisition of IAA that has been secreted by soil bacteria and is important in determining whether bacterial IAA stimulates or suppresses plant growth. Microbial IAA could be involved in the growth stimulation observed in our greenhouse assay. Production of plant growth regulators by the microorganisms is another important mechanism often associated with growth stimulation (Vessey, 2003). The balance between vegetative and reproductive growth is controlled by hormone signaling within the plant and therefore be highly influenced by it (Taiz and Zeiger, 1991). At relatively high concentrations, natural auxins, such as IAA, stimulate shoot elongation and root induction while reducing root elongation (Tanimoto, 2005; Gravel *et al.*, 2007) in his results, reported that *P. putida* subgroup B strain 1 and *T. atroviride* have the ability to synthesize IAA from different precursors in vitro, which supports the theory that microbial IAA could be involved in the growth stimulation observed in our greenhouse assay In the present investigation we tried to correlate the siderophore producing ability and Indole acetic acid (IAA), PHB production and inorganic phosphate solubilization ability of Fluorescent *Pseudomonas* with plant growth promoting ability. It was observed that a group of fluorescent *Pseudomonas* isolates induced significant growth effects on root and shoot on different crop plants, some only induce significant growth effects on shoot and or only root. Rice (*Oryza sativa* var. swarna): Efficacy of different isolates of *Pseudomonas* for rice plants varied to induce root and shoot length ranging from 7.15 to16.00 cm and 19.03 to 24.95 cm respectively. Maximum root length (16 cm) and shoot length (24.95 cm) were recorded when seeds were treated with P247 and P176 respectively (Table 7). Seed treated with isolate P247 has 6.67 cm (41.69%) more root length and isolate P176 has 4.03 cm (16.56%) shoot length as compared to control. Seven isolates were able to exert plant growth promoting activity on rice. Isolates P151, P233 significantly increased the root and shoot growth of rice. Isolates P85, P176, P216 significantly increased the shoot growth of rice. Isolates P67 and P247 significantly increased the root growth of rice. Wheat (*Triticum aestivum* var. GW-272): Efficacy of different isolates of *Pseudomonas* for wheat plants varied to induce root and shoot length ranging from 24.38 to 36.52cm and 30.77 to 39.78 cm respectively. Maximum root length (36.52cm) and shoot length (39.78 cm) were recorded when seeds were treated with P124 and P141 respectively as compared to control (Table 7). Seed treated with isolate P124 has 6.32 cm (17.30%) more root length and isolate P141 has 8.05 cm (20.23%) shoot length as compared to control. Twenty one isolates were

able to exert plant growth promoting activity on wheat. Isolates P124 significantly increased the root and shoot growth of wheat. Isolates P67, P72, P76, P85, P99, P126, P129, P141, P143, P151, P161, P167, P176, P179, P201, P205, P216, P233, P247 and P248 significantly increased the shoot growth. Bottlegourd (*Lagenaria siceraria*): The screening of *Pseudomonas spp*. treated seed of bottle gourd was evaluated 40 days after sowing. The efficacy of different species of *Pseudomonas* isolates varied to induce root length ranged from 20.33 to 112.73 cm for which P248 (112.73cm) isolate measured longest root length and P151(20.33cm) isolate shortest root length (Table 8). The shoot length measured ranged from 3.95 to 63.75 cm for which fluorescent *Pseudomonas spp.* isolate P167 (63.75cm) measured longest shoot length and P126 (3.95cm) isolate measured shortest shoot length).

Seed treated with isolate P248 has 61.05 cm (68.83%) more root length and isolate P167 has 46.15 cm (72.39%) shoot length as compared to control Thirteen isolates were able to exert plant growth promoting activity on bottle gourd. Seed biopriming of bottlegourd seeds with isolate P76, P99, P124 and P167increased the root and shoot length whereas biopriming with isolates P85, P126, P143, P176, P248 resulted only increase in shoot length. Seed biopriming of bottlegourd seeds with P72, P129, P141 and P151 increased the root length. Lathyrus (*Lathyrus sativus* var. KH-014): Efficacy of different isolates of *Pseudomonas* for lathyrus plants varied to induce root and shoot length ranging from12.79 to 25.70cm and 13.12 to 20.82cm respectively. Maximum root length (25.7cm) and shoot length (20.815 cm) were recorded when seeds were treated with P216 and P85 respectively (Table 8). Seed treated with isolate P216 has 7.51 cm (29.22%) more root length and isolate P85 has 7.45 cm (35.78%) shoot length as compared to control. Seventeen isolates were able to exert plant growth promoting activity on Lathyrus except P216 all the sixteen isolates (P5, P6, P72, P85, P99, P11, P124, P126, P129, P151, P167, P176, P201, P205, P247, P248) significantly promoted the shoot growth of lathyrus. Chickpea (*Cicer arientinum*): Efficacy of different isolates of *Pseudomonas* for chickpea plants varied to induce root and shoot length ranging from 16.75 to 38.85 cm and14.933 to 25.7 cm respectively. Maximum root length (38.85 cm) and shoot length (25.7 cm) were recorded when seeds were treated with P72 as compared to control (Table 8). Seed treated with isolate P72 had 10.52 cm (27.08%) and 9.37 cm (36.45%) more root and shoot length respectively as compared to control. Twenty isolates were able to exert plant growth promoting activity on chickpea. Isolate P72 significantly increase the root and shooth growth of chickpea whereas nineteen isolates P67, P76, P85, P99, P11, P124, P126, P129, P141, P143, P151, P161, P176, P201, P205, P216, P233, P247 significantly increase the shoot growth of chickpea. Greengram (*Vigna radiata* var. puspa vishal): Efficacy of different isolates of *Pseudomonas* for greengram plants varied to induce root and shoot length ranging from 13.99 to 22.15 cm and 13.7 to 20.385 cm respectively. Maximum root length (19.59cm) and shoot length (20.39 cm) were recorded when seeds were treated with P151 and P11 respectively (Table 9). Seed treated with isolate P11 has 6.61cm (32.81%) more shoot length as compared to control. Isolates P6, P67, P99, P11, P124, P129, P143, P151, P167, P176, P201, P216, P233 and P247 on biopriming greengram seeds promoted shoot growth whereas P141 promoted only increased root and shoot development. Blackgram (*Vigna mungo* var. T-U-94-2): Efficacy of different isolates of *Pseudomonas* for blackgram plants varied to induce root and shoot length ranging from 14.59 to 24.83 cm and 12.34 to18.62 cm respectively.

Table.1 Spectrophotometric determination of Indole acetic acid (IAA), PHB production and inorganic phosphate solubilization in different proportions by fluorescent *Pseudomonas* isolates

S. No.	Isolates	IAA production (µg/ml)	PHB Production (mg/ml)	Phosphate solubilization in Pikovskaya's		
				Broth (µg/ml)	Agar medium	
					with BCP	without BCP
1	P6	8.09	2.67	264	++	++
2	P11	13.36	5.37	407	+	++
3	P5	15.36	6.86	379	++	+
4	P201	22.27	6.86	518	++	+
5	P67	13.91	7.56	550	+++	+++
6	P124	63.18	7.82	356	+++	+++
7	P85	9.09	8.99	399	+	+++
8	P143	12.55	10.48	135	++	+
9	P141	10.27	11.55	383	+++	++
10	P205	21.91	11.83	347	++	+
11	P216	9.91	11.93	768	+++	+++
12	P167	9.64	11.97	95	++	+
13	P247	13	12.52	278	++	++
14	P76	11.09	12.91	484	+	++
15	P72	24	13.19	502	+	+
16	P248	14.36	13.24	335	+	+
17	P129	21	13.4	306	++	++
18	P176	12.36	13.9	194	+	++
19	P179	13.27	14.05	239	++	++
20	P151	13.45	14.25	88	+	+
21	P233	8.82	14.4	139	++	++
22	P161	16.18	14.52	455	+	+
23	P99	27.09	15.31	401	+++	+++
24	P126	13.64	15.74	365	+	+
Max.		63.18	15.74	768		
Min.		8.09	2.67	88		
Control				84		

+++: luxuriant/high phosphate solubilization; ++: medium phosphate solubilization; +: low phosphate solubilization; -: no phosphate solubilization.

Table.2 Fluorescent *Pseudomonas* isolates influencing root length in different crop plants

Treatment	Root length (cm)						
	Blackgram	Bottlegourd	Chickpea	Greengram	Lathyrus	Rice	Wheat
C	16.435ef±0.733	43.9efg±12.27	28.333bcdef±0.726	17.28bcde±2.10	18.185bcdefg±0.679	9.325efg±1.135	30.2bcdefg±0.742
P5	18.565bcdef±1.115	45.7defg±6.74	29bcdef±6.449	15.39de±2.58	20.565abcde±0.957	12.2abcdef±1.301	28.42defgh±1.369
P6	20.985abcd±1.401	42.83efgh±0.82	20.533fg±0.088	18.04bcd±1.80	19.415abcde±0.853	10.25cdefg±0.487	27.68efgh±1.685
P67	20.215bcde±1.540	54cdefg±8.01	27.333cdef±1.202	14.72de±0.48	19.385bcdef±2.496	14.5ab±2.480	27.24fgh±1.259
P72	20.225bcde±2.244	83.3b±1.15	38.85a±6.438	17.47bcde±1.12	19.085bcdef±1.464	10defg±0.970	27.56fgh±1.755
P76	19.15bcde±1.187	70.3bc±2.77	36.833ab±2.920	13.99e±2.29	12.785g±0.585	8.525fg±0.581	30.66bcdefg±0.897
P85	20.665abcde±0.959	41.83fgh±12.44	28.833bcdef±0.833	16.22cde±0.68	18.285bcdef±2.483	12.175abcdef±1.035	27.19gh±1.119
P99	19.765bcde±0.251	70.8bc±2.64	34.166abc±2.351	15.82cde±1.29	22.35ab±3.126	9.2efg±0.426	27.69efgh±1.102
P11	19.925bcde±1.858	44.5efg±6.01	29.833bcde±0.441	18.03bcd±0.44	22.575ab±1.424	8.5fg±0.951	31.86bcdef±1.999
P124	18.635bcdef±0.884	87.7b±7.23	26.833cdef±4.086	17.95bcd±0.77	21.375abcd±3.196	10.3cdefg±2.358	36.52a±2.766
P126	17.425def±0.574	64.95bcdef±8.54	23defg±2.021	15.99cde±1.49	16.375defg±1.830	9.775efg±0.559	30.07bcdefg±1.316
P129	19.715bcde±1.178	69.88bc±9.66	27.666cdef±0.882	15.69cde±2.80	16.465defg±1.723	10.825bcdefg±0.409	28.12efgh±1.907
P141	19.65bcde±0.991	67.83bcd±11.11	33.166abc±2.242	22.15a±0.76	15.665efg±2.070	7.15g±0.891	26.56gh±1.240
P143	22.135abc±2.148	59.18cdefg±4.64	33.65abc±3.839	18.49abcd±0.84	17.55bcdefg±0.484	7.625g±1.004	31.14bcdefg±1.775
P151	16.465ef±0.802	75.63bc±6.82	31.433abcd±2.599	19.59abc±1.48	13.965fg±0.380	13.625abcd±2.755	30.86bcdefg±1.150
P161	16.785def±1.574	42.4efgh±9.24	30.5abcde±2.180	15.94cde±0.38	15.775efg±1.730	12.5abcde±0.631	32.28abcde±1.513
P167	17.835def±1.218	73.5bc±16.98	22efg±4.726	18.63abcd±1.27	18.735bcdef±0.671	9.8defg±0.715	24.38h±1.906
P176	22.7ab±2.257	65.6bcde±10.00	24.25defg±3.320	17.08bcde±0.23	20.525abcde±2.771	10defg±1.772	34.51ab±1.602
P179	17.75def±2.027	20.33h±1.13	16.75g±1.299	20.37ab±0.17	16.7cdefg±1.198	7.8g±0.356	28.61defgh±1.749
P201	19.885bcde±1.499	52.88cdefg±7.83	24.4defg±3.114	16.47bcde±1.08	19.915bcde±3.689	8.625fg±2.138	29.44cdefg±1.610
P205	16.95def±1.551	42.7efgh±5.66	23.933defg±0.788	16.60bcde±1.80	18.615bcdef±0.742	7.55g±0.144	29.75cdefg±1.447
P216	19.675bcde±2.523	60cdefg±9.37	29.866bcde±4.332	18.00bcd±1.41	25.7a±2.957	8.7efg±1.079	27.41fgh±1.529
P233	24.825a±2.510	41.58gh±4.30	28.666bcdef±0.441	17.84bcde±1.07	15.715efg±1.047	13.9abc±2.200	32.82abcd±1.641
P247	18.125cdef±1.196	44.5efg±11.85	31.4abcd±2.358	16.00cde±0.97	19.315bcdef±1.818	16a±1.567	33.79abc±3.066
P248	14.585f±0.716	112.73a±4.10	26.033cdef±2.338	16.09cde±1.12	21.93abc±2.049	10defg±0.799	29.48cdefg±1.389
Max.	24.825a±2.510	112.73a±4.10	38.85a±6.438	22.15a±0.76	25.7a±2.957	16a±1.567	36.52a±2.766
Min.	14.585f±0.716	20.33h±1.13	16.75g±1.299	13.99e±2.29	12.785g±0.585	7.15g±0.891	24.38h±1.906
CD 0.01	5.7	30.907	11.443	-	7.243	5.077	6.165
CD 0.05	4.298	23.295	8.584	3.923	5.467	3.825	4.652
C V	15.917	27.961	18.49	16.217	20.76	26.234	12.461
F.cal	2.115	5.559	2.835	1.688	2.274	2.987	2.814

Table.3 Fluorescent *Pseudomonas* isolates influencing shoot length in different crop plants

Isolate #	Shoot length (cm)						
	Blackgram	Bottlegourd	Chickpea	Greengram	Lathyrus	Rice	Wheat
C	12.625mnop±0.3637	17.6efgh±1.254	16.333mn±0.498	13.7h±1.747	13.365i±1.094	20.3hijk±1.498	31.73jk±0.4107
P5	15.815defgh±0.3044	14.33fgh±0.794	14.933n±0.536	14.665fgh±0.830	16.635cdefg±0.835	20.775ghijk±0.771	33.04ij±0.6838
P6	18.025ab±0.5456	16.4efgh±0.589	21.066defghi±0.636	18.225abcd±1.19	19.135abc±0.382	21.65fghij±1.713	30.77k±1.0157
P67	13.375jklmnop±0.742	12.75fgh±0.777	21.5cdefg±1.510	17.625bcde±0.468	13.885hi±0.922	23.225abcdefg±0.390	36.04efg±0.5142
P72	13.165lmnop±0.4625	15fgh±1.780	25.7a±0.115	16.1defgh±1.022	16.2defgh±0.450	19.25jk±0.811	36.35defg±0.9332
P76	14.575hijk±0.3172	36.73bcd±5.981	21.266defgh±0.504	14.8fgh±0.452	13.15i±0.157	21.85efghi±0.835	36.75cdef±0.8353
P85	16.8bcde±0.3007	40.68bc±7.366	23.233bcd±0.536	14.985fgh±1.236	20.815a±0.780	24.45abcd±0.362	37.25bcde±0.8825
P99	18.615a±0.3912	56.73a±5.496	21.7cdefg±0.751	19.125abc±0.859	17.765bcdef±0.677	23.6abcdef±0.942	38.84ab±0.4621
P11	16defg±0.3969	11.1fgh±0.733	22.866bcde±0.754	20.385a±0.987	19.835ab±0.521	21.225fghijk±0.684	33.78hi±0.6555
P124	15.235fghi±0.6759	58.93a±9.433	23.833abc±0.167	17.615bcde±0.750	20.2ab±0.774	22.775abcdefgh±0.536	38.18abcd±1.1289
P126	12.5nop±0.3048	50.65ab±12.99	20.75efghi±1.010	15.325efgh±0.820	18.1bcde±0.174	22.35bcdefghi±0.598	36.89bcdef±0.8538
P129	14.025ijklm±0.4498	24.53def±4.514	25.066ab±0.924	16.865cdefg±1.170	18.015bcde±0.923	20.135ijk±0.613	38.55abc±0.5677
P141	17.515abc±0.4502	11.4fgh±1.023	18.933hijkl±0.470	16.4defg±0.524	14.8ghi±1.093	22.15cdefghi±0.366	39.78a±0.9069
P143	17.685abc±0.4719	37.4bcd±3.447	21defghi±1.732	19.175abc±1.301	15.285fghi±0.739	19.025k±1.424	35.9efg±0.2994
P151	12.335op±0.4575	20.85efg±0.296	21.066defghi±0.581	20.235a±1.473	16.8cdefg±1.365	24.725ab±0.201	35.58efgh±0.6486
P161	13.275klmnop±0.2087	12.58fgh±0.862	22.166cdef±0.928	16.185defg±0.648	13.115i±0.984	23.325abcdefg±0.239	35.83efg±0.4839
P167	13.885ijklmn±0.3319	63.75a±5.883	16.833lmn±0.899	16.465defg±0.390	16.85cdefg±1.646	23.15abcdefg±0.744	34.45ghi±0.4381
P176	17.165bcd±0.6932	37.5bcd±11.76	19.5ghijk±1.155	19.675ab±0.338	18.785abcd±1.573	24.95a±0.833	38.27abcd±0.6350
P179	16.5cdef±0.3857	3.95h±0.466	18.25jklm±0.722	16.065defgh±0.228	13.365i±0.546	22defghi±0.082	36.09efg±0.3703
P201	16.75bcde±0.4392	15.4fgh±1.362	19.733ghijk±0.536	16.935cdef±0.388	16.065efgh±0.933	21.4fghijk±1.485	38.59abc±0.9865
P205	14.175ijkl±0.3099	14.175fgh±0.312	20.166fghij±0.601	14.45gh±0.149	16.525defg±0.545	22.325bcdefghi±0.725	36.81cdef±0.3945
P216	14.7ghij±0.8147	11.55fgh±0.689	20.766efghi±0.865	16.965cdef±0.889	15.185fghi±0.508	24.675abc±1.071	35.16fgh±0.2973
P233	15.585efgh±0.8166	30.98cde±12.056	22.5cdef±1.155	18.065abcd±0.268	14.65ghi±0.899	24.325abcde±0.531	35.86efg±0.3638
P247	15.7efgh±0.4449	7.14gh±0.865	18.733ijkl±0.617	16.65defg±0.388	19.825ab±1.567	22.3bcdefghi±1.219	37.28bcde±0.4847
P248	16.8bcde±0.6334	36.7bcd±3.604	17.366klm±0.857	15.2efgh±0.228	20.705a±0.755	20.825ghijk±1.314	38.12abcd±0.5951
Max.	18.615a±0.3912	63.75a±5.883	25.7a±0.115	20.385a±0.987	20.815a±0.780	24.95a±0.833	39.78a±0.9069
Min.	12.335op±0.4575	3.95h±0.466	14.933n±0.536	14.665fgh±0.830	13.115i±0.984	19.025k±1.424	30.77k±1.0157
CD 0.01	1.857	20.522	3.19	3.268	3.447	3.39	2.636
CD 0.05	1.401	15.464	2.391	2.464	2.59	2.568	1.987
C V	6.495	41.673	7.075	10.361	10.994	8.167	4.372
F.cal	14.069	10.136	9.781	4.488	7.27	3.412	9.65

Table.4 Fluorescent *Pseudomonas* isolates inducing significant increase in root and shoot length in different crop plants and their Indole acetic acid (IAA), PHB production and inorganic phosphate solubilization ability

S. No.	Isolates	Increase in root and shoot growth	IAA production (µg/ml)	PHB Production (mg/ml)	Phosphate solubilization in Pikovskaya's		
					Broth (µg/ml)	Agar medium	
						with BCP	without BCP
1	P6	Blackgram	8.09	2.67	264	++	++
2	P72	Chickpea	24	13.19	502	+	+
3	P76	Bottlegourd	11.09	12.91	484	+	++
4	P99	Bottlegourd	27.09	15.31	401	+++	+++
5	P124	Bottlegourd, Wheat	63.18	7.82	356	+++	+++
6	P141	Greengram	10.27	11.55	383	+++	++
7	P143	Blackgram	12.55	10.48	135	++	+
8	P151	Rice	13.45	14.25	88	+	+
9	P167	Bottlegourd	9.64	11.97	95	++	+
10	P176	Blackgram	12.36	13.9	194	+	++
11	P233	Blackgram, Rice	8.82	14.4	139	++	++

Table.5 Fluorescent *Pseudomonas* isolates inducing significant increase development of shoot length in different crop plants and their Indole acetic acid (IAA), PHB production and inorganic phosphate solubilization ability

S. No.	Isolates	Increase in shoot growth	IAA production (µg/ml)	PHB Production (mg/ml)	Phosphate solubilization in Pikovskaya's		
					Broth (µg/ml)	Agar medium	
						with BCP	without BCP
1	P5	Blackgram, Lathyrus	15.36	6.86	379	++	+
2	P72	Lathyrus, Wheat	24	13.19	502	+	+
3	P161	Chickpea, Wheat	16.18	14.52	455	+	+
4	P179	Blackgram, Wheat	13.27	14.05	239	++	++
5	P6	Chickpea, Greengram, Lathyrus	8.09	2.67	264	++	++
6	P67	Chickpea, Greengram, Wheat	13.91	7.56	550	+++	+++
7	P76	Blackgram, Chickpea, Wheat	11.09	12.91	484	+	++
8	P141	Blackgram, Chickpea, Wheat	10.27	11.55	383	+++	++
9	P167	Greengram, Lathyrus, Wheat	9.64	11.97	95	++	+
10	P233	Chickpea, Greengram, Wheat	8.82	14.4	139	++	++
11	P11	Blackgram, Chickpea, Greengram, Lathyrus	13.36	5.37	407	+	++
12	P124	Blackgram, Chickpea, Greengram, Lathyrus	63.18	7.82	356	+++	+++
13	P126	Bottlegourd, Chickpea, Lathyrus, Wheat	13.64	15.74	365	+	+
14	P129	Chickpea, Greengram, Lathyrus, Wheat	21	13.4	306	++	++
15	P143	Bottlegourd, Chickpea, Greengram, Wheat	12.55	10.48	135	++	+
16	P151	Chickpea, Greengram, Lathyrus, Wheat;	13.45	14.25	88	+	+
17	P205	Blackgram, Chickpea, Lathyrus, Wheat;	21.91	11.83	347	++	+
18	P248	Blackgram, Bottlegourd, Lathyrus, Wheat	14.36	13.24	335	+	+
19	P99	Blackgram, Chickpea, Greengram, Lathyrus, Wheat;	27.09	15.31	401	+++	+++
20	P201	Blackgram, Chickpea, Greengram, Lathyrus, Wheat	22.27	6.86	518	++	+
21	P216	Blackgram, Chickpea, Greengram, Rice, Wheat	9.91	11.93	768	+++	+++
22	P247	Blackgram, Chickpea, Greengram, Lathyrus, Wheat	13	12.52	278	++	++
23	P85	Blackgram, Bottlegourd, Chickpea, Lathyrus, Rice, Wheat	9.09	8.99	399	+	+++
24	P176	Bottlegourd, Chickpea, Greengram, Lathyrus, Rice, Wheat	12.36	13.9	194	+	++

Table.6 Fluorescent *Pseudomonas* isolates inducing significant increase in root growth in different crop plants and their Indole acetic acid (IAA), PHB production and inorganic phosphate solubilization ability

S. No.	Isolates	Increase in root growth	IAA production (µg/ml)	PHB Production (mg/ml)	Phosphate solubilization in Pikovskaya's		
					Broth (µg/ml)	Agar medium	
						with BCP	without BCP
1	P67	Rice	13.91	7.56	550	+++	+++
2	P72	Bottlegourd	24	13.19	502	+	+
3	P129	Bottlegourd	21	13.4	306	++	++
4	P141	Bottlegourd	10.27	11.55	383	+++	++
5	P151	Bottlegourd	13.45	14.25	88	+	+
6	P216	Lathyrus	9.91	11.93	768	+++	+++
7	P247	Rice	13	12.52	278	++	++

Table.7 Fluorescent *Pseudomonas* isolates inducing significant reduction in root and shoot length in different crop plants and their Indole acetic acid (IAA), PHB production and inorganic phosphate solubilization ability

S. No.	Isolates	Inhibitory effects on root & shoot development	IAA production (µg/ml)	PHB Production (mg/ml)	Phosphate solubilization in Pikovskaya's		
					Broth (µg /ml)	Agar medium	
						with BCP	without BCP
1	P143	Rice	12.55	10.48	135	++	+
2	P76	Lathyrus	11.09	12.91	484	+	++
3	P6	Wheat, Bottlegourd	8.09	2.67	264	++	++
4	P161	Bottlegourd	16.18	14.52	455	+	+
5	P179	Bottlegourd	13.27	14.05	239	++	++
6	P205	Bottlegourd	21.91	11.83	347	++	+

Table.8 Fluorescent *Pseudomonas* isolates inducing significant reduction in root length in different crop plants and their Indole acetic acid (IAA), PHB production and inorganic phosphate solubilization ability

S. No.	Isolates	Inhibitory effects on root development	IAA production (µg/ml)	PHB Production (mg/ml)	Phosphate solubilization in Pikovskaya's		
					Broth (µg/ml)	Agar medium	
						with BCP	without BCP
1	P72	-	24	13.19	502	+	+
2	P11	Rice	13.36	5.37	407	+	++
3	P216	Rice;	9.91	11.93	768	+++	+++
4	P5	Greengram	15.36	6.86	379	++	+
5	P6	Chickpea	8.09	2.67	264	++	++
6	P124	Chickpea	63.18	7.82	356	+++	+++
7	P143	Lathyrus	12.55	10.48	135	++	+
8	P151	Lathyrus	13.45	14.25	88	+	+
9	P247	Greengram	13	12.52	278	++	++
10	P161	Greengram	16.18	14.52	455	+	+
11	P99	Greengram, Rice	27.09	15.31	401	+++	+++
12	P76	Greengram, Rice	11.09	12.91	484	+	++
13	P141	Lathyrus, Rice	10.27	11.55	383	+++	++
14	P67	Chickpea, Greengram	13.91	7.56	550	+++	+++
15	P176	Chickpea, Greengram	12.36	13.9	194	+	++
16	P85	Bottlegourd, Greengram	9.09	8.99	399	+	+++
17	P233	Bottlegourd, Lathyrus	8.82	14.4	139	++	++
18	P179	Chickpea, Lathyrus, Rice	13.27	14.05	239	++	++
19	P201	Chickpea, Greengram, Rice	22.27	6.86	518	++	+
20	P205	Chickpea, Greengram, Rice	21.91	11.83	347	++	+
21	P126	Chickpea, Greengram, Lathyrus	13.64	15.74	365	+	+
22	P129	Chickpea, Greengram, Lathyrus;	21	13.4	306	++	++
23	P167	Bottlegourd, Greengram, Lathyrus	9.64	11.97	95	++	+
24	P248	Blackgram, Chickpea, Greengram	14.36	13.24	335	+	+

Table.9 Fluorescent *Pseudomonas* isolates inducing significant reduction in shoot length in different crop plants and their Indole acetic acid (IAA), PHB production and inorganic phosphate solubilization ability

S. No.	Isolates	Inhibitory effects on shoot development	IAA production (µg/ml)	PHB Production (mg/ml)	Phosphate solubilization in Pikovskaya's		
					Broth (µg/ml)	Agar medium	
						with BCP	without BCP
1	P85	Wheat	09.09	8.99	399	+	+++
2	P99	Wheat	27.09	15.31	401	+++	+++
3	P179	Wheat	13.27	14.05	239	++	++
4	P205	Wheat;	21.91	11.83	347	++	+
5	P248	Wheat	14.36	13.24	335	+	+
6	P151	Blackgram;	13.45	14.25	88	+	+
7	P247	Bottlegourd	13	12.52	278	++	++
8	P11	Rice, Bottlegourd	13.36	5.37	407	+	++
9	P129	Rice, Wheat	21	13.4	306	++	++
10	P67	Bottlegourd, Wheat	13.91	7.56	550	+++	+++
11	P126	Blackgram, Wheat;	13.64	15.74	365	+	+
12	P141	Bottlegourd, Wheat;	10.27	11.55	383	+++	++
13	P201	Bottlegourd, Wheat	22.27	6.86	518	++	+
14	P216	Bottlegourd, Wheat	09.91	11.93	768	+++	+++
15	P72	Bottlegourd, Rice, Wheat	24	13.19	502	+	+

Maximum root length (24.83 cm) and shoot length (18.62 cm) were recorded when seeds were treated with P233 and P99 respectively (Table 9). Seed treated with isolate P233 has 8.39 cm (33.8%) more root length and isolate P99 has 5.99 cm (32.16%) shoot length as compared to control. Fourteen isolates expressed plant growth promoting activity on blackgram. Isolates P6, P143, P176 and P233 were able to promote significantly root and shoot length, whereas isolates P5, P76, P85, P99, P11, P124, P141, P179, P201, P205, P216, P247, P248 promote significantly root length of blackgram.

Fluorescent *Pseudomonas* isolates inducing significant increase in root and shoot growth in different crop plants

Increase in root and shoot length in one crop: P72 Chickpea; P141 Greengram; P151 Rice. Increase in root and shoot length in two crops: P233 Blackgram, Rice; P124 Bottlegourd, Wheat. Three fluorescent *Pseudomonas* isolates increased the root and shoot length of the same crop: P6, P143, P176 (Blackgram); P76, P99, P167 (Bottlegourd).

Isolates inducing significant increase in shoot growth in different crop plants

It was observed that Fluorescent *Pseudomonas* isolates the frequency of inducing shoot length was more as compared to root length.

In the order of decreasing frequency of fluorescent *Pseudomonas* isolates inducing increased shoot growth in different crops is as follows: Wheat (20)> Chickpea (19)> Lathyrus (16)> Greengram (14)> Blackgram (13)> Bottlegourd (5)> Rice (3).

Fluorescent *Pseudomonas* isolates P5, P6, P11, P124 did not increased the shoot length in rice or wheat. Among cereals wheat was

more responsive and expressed increased shoot length with 20 different Fluorescement *Pseudomonas* isolates whereas rice expressed increased shoot length with only three isolates P216, P85 and P176.

Bottlegourd are very responsive to IAA but expressed increased shoot length with only P126, P143, P248. Fluorescent *Pseudomonas* isolate P124 was the highest IAA producer but was not able to induce increased root or shoot length in bottlegourd.

Fluorescent *Pseudomonas* isolates increased the shoot length only in one legume crop P161 Chickpea; P72 Lathyrus; P179 Blackgram

Fluorescent *Pseudomonas* isolates increased the shoot length in any two legume crop P5, P248 Blackgram, Lathyrus; P167 Greengram, Lathyrus; P126 Chickpea, Lathyrus; P67, P143, P233 Chickpea, Greengram; P76, P141 Blackgram, Chickpea.

Fluorescent *Pseudomonas* isolates increased the shoot length in three legumes crop P6, P129, P151, P176 Chickpea, Greengram, Lathyrus; P85, P205 Blackgram, Chickpea, Lathyrus; P216 Blackgram, Chickpea, Greengram

Fluorescent *Pseudomonas* isolates increase the shoot length in all four legumes (Blackgram, Chickpea, Greengram, Lathyrus) P11, P124, P99, P201, P247.

Isolates inducing significant increase in root growth in different crop plants

The frequency of Fluorescent *Pseudomonas* isolates inducing increased root length was four (P72, P129, P141 and P151) whereas only two P67 and P247 induced increased root length in rice and only one in lathyrus by P216.

PGPR can alter root architecture and promote plant development with the production of different phytohormones like IAA, gibberellic acid and cytokinins (Kloepper *et al.,* 2007). Similarly, significant shoot growths in maize and rice dwarf mutants were promoted by gibberellins-like substances excreted by *Azospirillum* spp. (Boiero *et al.,* 2007). IAA-mediated ethylene production could increase root biomass, root hair number and consequently the root surface area of PGPR inoculated tomato plants (Ribaudo *et al.,* 2006). Involvement of PGPR formulated cytokinins were also observed in root initiation, cell division, cell enlargement and increase in root surface area of crop plants through enhanced formation of lateral and adventitious roots(Werner *et al.,* 2003). Recently, it has been established that the working pathways of these phytostimulators leading to overall development in crop plants are differently regulated by catabolite repression (Zaidi *et al.,* 2009) as physiological regulator of biofilm formation.IAA biosynthesis has been correlated with stimulation of root proliferation by rhizosphere bacteria (Persello-Cartieaux *et al.,* 2003; Spaepen *et al.,* 2007), which enhanced uptake of nutrients by the associated plants (Lifshitz *et al.,* 1987). Moreover, inoculation with an *Azospirillum brasilense* Sp245 mutant strain, strongly reduced in auxin biosynthesis or addition of increasing concentrations of exogenous auxin to the plant growth medium, indicated that the differential response to *A. brasilense* Sp245 among the common bean (*Phaseolus vulgaris* L.) genotypes is related to the bacterial produced auxin (Remans *et al.,* 2008). IAA affects plant cell division, extension, and differentiation; stimulates seed and tuber germination; increases the rate of xylem and root development; controls processes of vegetative growth; initiates lateral and adventitious root formation; mediates responses to light, gravity and florescence;

affects photosynthesis, pigment formation, biosynthesis of various metabolites, and resistance to stressful conditions (Tsavkelova *et al.,* 2006; Spaepen and Vanderleyden 2011). IAA synthesized by bacteria may be involved at different levels in plant-bacterial interactions. In particular, plant growth promotion and root nodulation are both affected by IAA. The role of IAA that was synthesized by the PGPB *Pseudomonas putida* GR12-2 in the development of canola roots was studied following the construction of an IAA-deficient mutant of this strain (Patten and Glick 2002a). Seed inoculation with wild-type *P. putida* GR12-2 induced the formation of roots that were 35–50% longer than the roots from seeds treated with the IAA-deficient mutant and the roots from uninoculated seeds. On the other hand, inoculation of mung bean cuttings with a mutant of the same strain (Xie *et al.,* 1996), which overproduces IAA, yielded a much greater number of shorter roots compared with controls (Mayak *et al.,* 1999).

This result was explained by the combined effect of auxin on growth promotion and inhibition of root elongation by ethylene(Jackson 1991). The bacterial IAA that was incorporated by the plant stimulated the activity of the enzyme ACC synthase, resulting in increased synthesis of ACC (Jackson 1991), and a subsequent rise in ethylene that inhibited root elongation (Riov and Yang 1989). Overall, bacterial IAA increases root surface area and length, and thereby provides the plant has greater access to soil nutrients. In addition, bacterial IAA loosens plant cell walls and as a result facilitates an increasing amount of root exudation that provides additional nutrients to support the growth of rhizosphere bacteria.

It was observed that Fluorescent *Pseudomonas* isolates also reduced root shoot length as compared un-treated control.

Fluorescent *Pseudomonas* isolates inducing reduced root and shoot length in different crop plants

In the order of decreasing frequency of fluorescent *Pseudomonas* isolates inducing reduction in root and shoot length in Bottlegourd (4), Rice (1), Lathyrus (1) and Wheat (1)

Fluorescent *Pseudomonas* isolates inducing reduced root length in different crop plants

Except isolate P72 all the isolates induced reduction in root length. Because seed biopriming places the bacterium directly in contact with soil and after germination with rhizoplane / rhizosphere of the germinated seed, suggesting that the compatible / incompatible interaction between the host and bacterium might result into induced increased or decreased growth of the root. This might be the reason why the frequency of fluorescent *Pseudomonas* isolates inducing reduction in root length (23 out of 24 isolates) in different crop plants was very high as compared to isolates inducing reduction in shoot length (16 out of 24 isolates).

Fluorescent *Pseudomonas* isolates inducing reduced shoot length in different crop plants

In the order of decreasing frequency of fluorescent *Pseudomonas* isolates inducing reduction in shoot length in Wheat (12), Bottlegourd (7), Rice (3) Blackgram (2).

Inhibitory effect of some deleterious rhizobacteria through IAA secretion has been related to various bacterial species including *Enterobacter taylorae, Klebsiella planticola, Alcaligenes faecalis, Xanthomonas maltophila, Pseudomonas* sp. and *Flavobacterium* sp. (Sarwar and Kremer 1995; Suzuki *et al.,* 2003). Mutants of

Pseudomonas putida that produced high levels of IAA inhibited root growth of seedlings of canola (*Brassica campestris*) by ca. 33% (Xie *et al.*, 1996). Thus, ambiguity about effect of IAA on growth of root, shoot and rate of seedling emergence has been reported (Freitas and Germida 1990; Sarwar and Kremer 1995; Barazani and Friedman 1999) (Table 2–6). Despite the potential of allelopathic bacteria and growth-mediating allelochemicals in agriculture, it is one of the poorly understood areas of plantmicrobe interactions. Further work is needed to characterize bacteria and allelochemicals from the rhizosphere soil and to study their effect on the crop plants.

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