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Phosphate solubilizing and indole-3-acetic acid producing bacteria from the soil of Garhwal Himalaya aimed to improve the growth of rice

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Abstract

In the present study, soil bacteria from rainfed agriculture field of Garhwal Himalaya, just prior to sowing of summer crop, were isolated and initially tested for solubilization of inorganic phosphate, production of indole acetic acid (IAA) and siderophore. Two bacterial isolates, having efficient P- solubilizing activity in solid medium, were identified using 16S rRNA sequence analysis as *Pseudomonas koreensis* strainYB1 *Arthrobacter nitroguajacolicus* strainYB3 and three bacterial isolates, producing high amount of IAA in liquid medium, were identified as *Klebsiella oxytoca* strainYB2 and two strain of *Arthrobacter nitroguajacolicus*, strainYB4 and YB5, respectively. In culture medium supplemented with L-Tryptophan, *Klebsiella oxytoca* produced high amount of IAA (337.44µg l^{-1}). The selected five bacterial strains were further tested for tricalcium phosphate (TCP) solubilizing abilities at three different incubation temperature viz., 4ºC, 10ºC and 28ºC, under in vitro conditions. At 28ºC, three bacterial strains *Pseudomonas koreensis*, *Arthrobacter nitroguajacolicus* strainYB4 and *Klebsiella oxytoca* solubilized the phosphate efficiently. At 10ºC only two strains, *Pseudomonas koreensis* and *Arthrobacter nitroguajacolicus* strainYB4 solubilized phosphate efficiently as compared to other strains. These five bacterial strains were tested for nitrogen, catalase activity, starch and cellulose hydrolysis as well as growth promotion activity on rice, under controlled conditions. All the five bacterial strains efficiently increased the biomass and phosphorus uptake in Swarna and Swarna sub1 varieties of rice.

Key words

Indole acetic acid production, Phosphate solubilization, Plant growth promotion

Introduction

Microorganisms perform an important role in supplying nutrients to plants and reducing the demand of chemical fertilizers (Cakmakci *et al*., 2006). Plant growth promoting rhizobacteria (PGPR) enhance plant growth and productivity by producting of IAA and solubilization of phosphate (Datta *et al*., 2011). Phosphate solubilizing rhizobacteria solubilize phosphates, unavailable to the plants, in soil into available form and increase its availability for plant uptake (Gyaneshwar *et al*., 2002). In addition, PGPRs are directly involved in increased uptake of nitrogen, synthesis of phytohormones, solubilization of minerals, and production of siderophores that chelate iron and make it available to plant root and thus are involved in plant growth (Glick, 1995; Bowen and Rovira, 1999; Sivasakthi *et al.,* 2014). IAA production capacity is widespread among soil and plant associated bacteria (Patten and Glick, 1996) which play an important role in many aspects of root growth, development and differentiation (Aloni *et al*., 2006). Several studies have shown that bacteria isolated from soil can effectively be established as bioinoculant in the native plant environment as compared to non-native strains (Pandey *et al*., 1998).

The soils of Uttarakhand Himalaya are generally acidic in nature, low in moisture content, and organic matter. Applied water soluble phosphorus rapidly fixed to the unavailable form thus, accounting for low phosphate use efficiency (Pal, 1998). **S**oil sample from different altitudes in Uttarakhand Himalaya has been investigated for general, as well as, rhizospheric soil microbial diversity (Pandey *et al*., 2006a). Bacterial species, mostly

belonging to the genus *Bacillus* and *Pseudomonas* have been isolated and characterized from the rhizosphere of various plants of the Indian Himalayan Region (Negi *et al*., 2005; Pandey *et al*. 2006a; 2006b; 2011), yet the diversity of soil bacteria, which can be used as a bioinoculant for endogenous flora, remains elusive.

Rice is a usual staple food of the region (Mehta *et al*., 2010) and farmers prefer high yielding varieties of rice as compared to traditional varieties (Maikhuri *et al*., 2001). Swarna sub1 is a high yielding variety of rice developed by International Rice Research Institute (IRRI) by incorporating SUB1, a submergence tolerance gene, in popular rice variety Swarna (Anonymous, 2010), which is widely grown in drought-prone environments due to its high yield and other desirable traits (Mackill *et al*., 2010). The objective of the present study was to utilize the cultural and functional diversity of phosphate solubilizing and IAA producing bacteria from the soil of Garhwal Himalaya, for the improvement of growth of rice.

Materials and Methods

Isolation of bacteria : Bacterial strains were isolated from the soil samples collected from rainfed agriculture field of the Garhwal Himalayan region. Soil was collected during summer season before sowing the summer crop. Isolation of soil bacteria, by serial dilution was completed within 72 hr of sample collection. Bacteria present on nutrient agar (Himedia, India) plates were counted and then repeatedly streaked to purify.

Plant growth promoting (PGP) properties of bacterial isolates : Phosphate solubilization activities of selected bacterial isolates were measured (Pikovaskya, 1948) and solubilization index was determined.

Production of indole acetic acid (IAA) was calorimetrically determined (Gordon and Weber, 1951) by mixing 2 ml of Salkowski reagent (1ml of 0.5M $FeCl_3$ was dissolved in 50ml 35% perchloric acid) with 1 ml of supernatant from fresh cultures in medium containing (gl^{-1}): sodium glutamate 1g, sodium succenate 5g; $(NH_4)_2 SO_4$ 0.4g; $MgSO_4$ 0.2g; K_2HPO_4 0.5g. Pure indole-3-acetic acid (Sigma, USA) was used as standard. Each experiment was replicated three times. Chrome Azurol S (CAS) assay was used to detect siderophores produced by soil bacteria (Schwyn and Neilands, 1987).

Three bacterial isolates producing high amount of IAA, and two efficient phosphate solubilizing bacterial isolates were further evaluated for their biochemical and plant growth promoting (PGP) activity. Quantitative estimation of phosphate solubilization was carried out in 50 ml of modified Pikovaskiya broth containing ($g\ l^{-1}$): 10 g glucose; 5g $Ca_3(PO_4)_2$; 0.2 g KCl; 0.5 g $(NH_4)_2SO_4$; 0.2 g NaCl; 0.1 g $MgSO_4.7H_2O$; 0.002 g $MnSO_4.H_2O$; 0.002 g $FeSO_4.7H_2O$ and yeast extract 0.5g. The pH was adjusted to 7.0 before autoclaving. For inoculation, bacterial cells were grown up to 10^8-10^9c.f.u. ml^{-1} in nutrient broth. 100 µl of each bacterial culture was inoculated into 50 ml Pikovaskiya broth and incubated in incubator shaker (180 rev min^{-1}) at three different temperature *viz* 4, 10 and 28ºC. 10 ml culture was harvested every 24 hr interval for determination of pH and centrifugation at 6,000 rev min^{-1} for 15 min. Sterile uninoculated medium served as control. Remaining phosphate in the culture supernatant was estimated (Fiske and Subbarow, 1925) with modification. Culture supernatant (200µl) was mixed with 200 µl of 10% (w/v) trichloroacetic acid in a test tube to which was added 4 ml of color reagent (1:1:1:2 ratio of 3 M H_2SO_4 2.5% (w/v) Ammonium molybdate 10% (w/v) Ascorbic acid and distilled water) and incubated at room temperature (28 ± 2 ºC), for 15 min. The absorbance of the developing blue color was measured at 820 nm using spectrophotometer (RAY LEIGH, UV-2601). The amount of soluble phosphorus was detected from the standard curve of KH_2PO_4. All phosphate determinations were made in triplicate. All five bacterial isolates were also tested for nitrogen fixation (Okon *et al*., 1977), starch hydrolysis (Conn *et al*., 1957), cellulose hydrolysis (Apun *et al*., 2000) and catalse test by observing the evolution of oxygen when a loopful of each culture was mixed with 50 µl of 3% (v/v) H_2O_2 on a glass slide for 1 min.

Plant growth promotion assay : Pot experiment, with selected bacterial strains, was conducted using rice (*Oryza sativa* var. Swarna and Swarna sub1) as test species. Seeds were procured from IRRI New Delhi, India. Soil, for experiment, was taken from the agriculture field of G. B. Pant University of Agriculture and Technology, Pantnagar. pH of soil was 8.31 having 1.2% organic carbon, 186.7kg ha^{-1} nitrogen, 34.91kg ha^{-1} of phosphorus and 145.6 kg ha^{-1} potassium. Before filling the pot, the soil was steam sterilized after every alternate day for three days. Experiment was conducted in pots (500 ml capacity) with three replicates. Rice seeds were surface disinfected by immersion in 70% ethanol and 3% (v/v) sodium hypochlorite for 1 min and 5 min, respectively and left for germination in Petri dish under sterilized condition. Four equally germinated seeds were sown per pot and finally after a week, two were maintained per pot. Inoculation was carried out by adding 1 ml bacterial culture suspension having 10^7-10^8 cfu, in a hole created by pressing the germinated seed into it at the time of sowing. The pots were kept inside a greenhouse where temperature: 27±2°C, photo period: 16/8h day/night cycle, light intensity: 400$Em^{-2}s^{-1}$, (400-700 nm), and relative humidity: 60% were maintained 20 days of sowing, 10 ml of phosphorus free nutrient solution (Hoagland and Arnon, 1950) was supplied weekly to each pot. For evaluation of growth promotion effect of selected isolates, plants were uprooted 45 days after inoculation; height, fresh and dry weight of root and shoot were recorded. Dry weight of the sample was determined by placing the root and shoot samples separately into paper bags and drying them in an oven at 60ºC for 48 hr.

DNA extraction, 16S rRNA amplification and sequencing : DNA, from bacterial isolates, was extracted using CTAB method (Jaufeerally-Fakim and Dookun, 2000). The 16S rRNA genes of

bacteria were amplified by GM3f (5'-AGAGTTTGATCMTGGC-3') as a forward primer and GM4r (5'TACCTTGTTACGACTT-3') as a reverse primer (Muyzer et al. 1995). The 16S rRNA sequences of all five bacterial isolates were aligned with BLAST (Altschul *et al.,* 1990) algorithms to the genebank nucleotide database sequences (http://blast.ncbi. nih.gov/Blast.cgi).

Statistical analysis : Statistical analysis was performed with the Statistical Package for Social Sciences (SPSS) software, and treatment means were compared in Tukey HSD, at 1% level of significance.

Results and Discussion

On the basis of morphological characteristics, a total of 23 bacterial strains from soil were tested for their *in vitro* plant growth promoting (PGP) properties. The metabolic capacities of soil bacterial isolates assayed showed that out of 23 isolates, 19 isolates (82.61%) solubilized inorganic phosphate on PVK plate. The phosphate solubilization index of the isolates varied from 2.28 to 12.31. The isolate PGP1 exhibited the highest phosphate solubilization index followed by PGP9 (Table1). On the basis of 16S rRNA sequence analysis, isolate PGP1, showed close proximity with *Pseudomonas koreensis* strain Y17 with 99% similarity and isolate, PGP9 with *Arthrobacter nitroguajacolicus* strain TC1 with 100% similarity. 16S rRNA sequences of the bacterial isolates have been submitted in Gene Bank (NCBI) data base under accession number KC750201, KC750203. The objective of selecting the phosphate solubilizing bacteria (PSB) for growth promotion of rice is based on the fact that phosphate solubilization has received attention as it is considered as an important attribute of plant growth–promoting bacteria. PSB improve solubilization of unavailable soil phosphate resulting in high efficiency of phosphorus use (Sharma *et al*., 2013). In the present study, cultures released greater quantities of IAA in the presence of tryptophan a physiological precursor in culture medium. Out of 23 isolates, 21 (91.30%) were found positive for IAA. High amount of IAA production was observed in PGP4 (337.44 µg ml^{-1}) followed by PGP15 (220.77 µg ml^{-1}) and PGP16 (124.22 µg ml^{-1}). 16S rRNA sequence analysis of the isolate PGP4 showed close proximity with *Klebsiella oxytoca* strain SHD-1 with 99% similarity (accession number KC750202) and PGP15 and PGP16 with *Arthrobacter nitroguajacolicus* strain TC1 with 99% and 94% similarity (accession number KC750204 and KC750205), respectively. Bacteria producing IAA are considered as an effective tool for screening of growth promoting microorganisms, as many reports suggest that IAA producing bacteria have a profound effect on plant growth (Govindarajan *et al*., 2007; Ali and Hasnain, 2007). They promotes lateral and adventitious root formation, which can facilitate high root surface area for nutrient absorption from soil (Aloni *et al*., 2006). In the

Table 1 : Plant growth promoting properties of bacterial isolates

Isolates	Gram staining	Phosphate Solubilization Index(PVI)	IAA production (µg ml^{-1})	Siderophore production
PGP1	negative	12.33 ± 1.39^{a}	55.67 ± 10.40^{defg}	+++
PGP2	negative	5.63 ± 0.41^{d}	70.33 ± 10.02^{cdef}	+++
PGP3	negative	5.31 ± 0.31^{d}	ND	+++
PGP4	negative	5.99 ± 0.5^{d}	337.44 ± 58.58^{a}	+++
PGP5	negative	2.46 ± 0.2^{e}	10.00 ± 3.18^{fg}	+++
PGP6	negative	6.76 ± 0.2^{d}	41.78 ± 14.22^{defg}	-
PGP7	positive	ND	51.78 ± 17.80^{defg}	-
PGP8	negative	2.34 ± 0.12^{e}	39.78 ± 1.90^{defg}	++
PGP9	positive	9.17 ± 0.51^{b}	49.44 ± 7.34^{defg}	+++
PGP10	negative	2.68 ± 0.09^{e}	ND	++
PGP11	negative	ND	27.78 ± 1.39^{defg}	+++
PGP12	negative	6.68 ± 0.49^{d}	69.44 ± 21.53^{cdef}	+++
PGP13	negative	5.73 ± 0.38^{d}	20.78 ± 4.14^{efg}	++
PGP14	positive	3.05 ± 0.38^{e}	76.00 ± 4.06^{cde}	+
PGP15	positive	8.44 ± 0.66^{bc}	220.78 ± 40.78^{b}	++
PGP16	positive	6.86 ± 0.15^{cd}	124.23 ± 23.89^{c}	+
PGP17	negative	2.44 ± 0.09^{e}	56.33 ± 9.85^{defg}	+
PGP18	negative	ND	14.33 ± 3.21^{efg}	+
PGP19	negative	ND	19.00 ± 4.33^{efg}	+++
PGP20	negative	2.5 ± 0.2^{e}	19.67 ± 0.88^{efg}	++
PGP21	negative	6.32 ± 0.15^{d}	50.33 ± 9.39^{defg}	-
PGP22	negative	2.46 ± 0.08^{e}	20.00 ± 7.69^{efg}	+
PGP23	negative	6.52 ± 0.7^{d}	86.56 ± 4.86^{cd}	-

Results are mean ± SD of three analysis. Means with different superscripts significantly different from each other ($P < 0.01$); ND denotes not detected. – negative ,+ low, ++ medium, +++ high

present study, *Klebsiella oxytoca* showed much higher amount of IAA (337.44 µg ml^{-1}) as reported previously, where *Klebsiella* sp. produced 291.97 µg ml^{-1} IAA (Chaiharn and Lumyong, 2011) and *Klebsiella pneumonia* produced 135 µg ml^{-1} of IAA (Rueda-Pente *et al*., 2004). However, it has been reported that IAA production by PGP bacteria can vary among different species and strains, and it is also influenced by culture condition, growth stage and substrate availability (Mirza *et al*., 2001). In spite of being good producers of IAA, these bacteria were also positive for phosphate solubilization. Similarly, *Pseudomonas koreensis* and *Arthrobacter nitroguajacolicus* strainYB3 were found to be high phosphate solubilizer strain and have also produced IAA to some extent. Some findings suggested that phosphate solubilizing bacteria (PSB) can produce IAA (Jha and Kumar, 2009; Chaiharn and Lumyong, 2011). Similar to phosphate solubilization, 19 isolates (82.61%) were found positive for siderophore production activity by forming orange halos around the colonies after 12 days of incubation (Table 1). The result indicated that these isolates could be used against various pathogens (Negi *et al*., 2005).

It is believed that direct measurement of P-solubilization in broth assay is likely to give more reliable results than regular plate assay (Johri *et al*., 1999). However, these bacteria were isolated from high altitude region of the Indian Himalaya, where temperature fluctuation is a common phenomena. In this respect, quantitative estimation of phosphate solubilization were carried out, samples were taken at 24hr interval up to 3 days of incubation at 4, 10 and 28ºC, respectively. *Arthrobacter nitroguajacolicus* strainYB4 (98.52 µg ml^{-1}), *Pseudomonas koreensis* (98.5 µg ml^{-1}) and *Klebsiella oxytoca* (96.87 µg ml^{-1}) efficiently solubilized phosphate after three days of incubation at 28^{0}C. These strains significantly reduced the pH of medium as compared to two other strains (Table 2). At 4 and 10ºC, only *Pseudomonas koreensis* and *Arthrobacter nitroguajacolicus* strainYB4 showed marked phosphate solubilizing activities as well as reduced the pH of medium effectively, as compared to other strains (Table 2). Bacteria can produce organic acid as a result of glucose metabolism present in the broth, which may be considered to be responsible for the decline of pH of the broth (Hwangbo *et al*., 2003). However, decrease in pH of medium was found lesser when temperature were lowered from 28ºC to 4ºC. This might be due to lowering of functional activity of bacteria at low temperature. Bacteria that effectively decrease pH of medium during growth are considered as efficient P- solubilizers (Khan *et al*., 2006). In this respect, *Pseudomonas koreensis* and *Arthrobacter nitroguajacolicus* strainYB4 and *Klebsiella oxytoca* were found to be efficient P- solubilizers under optimal condition of growth and *Pseudomonas koreensis* and *Arthrobacter nitroguajacolicus* strainYB4 even under at low temperature of growth condition. Bacterial strain YB3 and YB4 did not show direct correlation of P- solubilization activity in solid and liquid media. They showed better P-solubilization activity in solid

Table 2 : Phosphate solubilizing activity of selected bacterial isolates at different temperatures

	Phosphate solubilization at 4 ºC (µg ml^{-1} ± SD)			pH change Phosphate solubilization at 4 ºC (pH ± SD)		
Strain	**24hr**	**48hr**	**72hr**	**24hr**	**48hr**	**72hr**
YB1	5.28 ± 0.09^{d}	9.46 ± 0.12^{c}	22.18 ± 0.31^{c}	6.14 ± 0.07^{a}	5.93 ± 0.04^{b}	5.71 ± 0.12^{b}
YB2	1.59 ± 0.09^{a}	2.13 ± 0.13^{a}	2.41 ± 0.09^{a}	6.61 ± 0.06^{a}	6.39 ± 0.08^{a}	6.28 ± 0.07^{a}
YB3	2.15 ± 0.11^{b}	2.49 ± 0.24^{a}	2.71 ± 0.18^{a}	6.49 ± 0.27^{a}	6.35 ± 0.05^{a}	6.2 ± 0.07^{a}
YB4	4.73 ± 0.09^{c}	8.9 ± 0.11^{b}	19.45 ± 0.11^{b}	6.26 ± 0.05^{a}	5.94 ± 0.04^{b}	5.77 ± 0.11^{b}
YB5	2.36 ± 0.09^{b}	2.6 ± 0.09^{a}	2.71 ± 0.12^{a}	6.60 ± 0.09^{a}	6.41 ± 0.09^{a}	6.24 ± 0.11^{a}
Phosphate solubilization at 10 ºC (µg ml^{-1} ± SD)				**pH change Phosphate solubilization at 10 ºC (pH ± SD)**		
isolates	**24hr**	**48hr**	**72hr**	**24hr**	**48hr**	**72hr**
YB1	12.57 ± 0.13^{a}	28.9 ± 0.56^{a}	95.34 ± 0.61^{a}	5.51 ± 0.04^{a}	4.75 ± 0.10^{c}	4.34 ± 0.17^{b}
YB2	10.85 ± 0.05^{b}	10.33 ± 0.15^{cd}	14 ± 0.26^{d}	5.65 ± 0.04^{a}	5.54 ± 0.04^{a}	5.54 ± 0.19^{a}
YB3	10.78 ± 0.13^{b}	9.63 ± 0.15^{d}	20.3 ± 0.2^{c}	5.65 ± 0.01^{a}	5.49 ± 0.09^{a}	5.3 ± 0.16^{a}
YB4	11.62 ± 0.03^{a}	19.19 ± 0.13^{b}	94.03 ± 0.2^{b}	5.6 ± 0.09^{a}	5 ± 0.02^{b}	4.25 ± 0.09^{b}
YB5	10.29 ± 0.9^{b}	11.25 ± 0.44^{c}	12.56 ± 0.23^{e}	5.73 ± 0.11^{a}	5.58 ± 0.02^{a}	5.55 ± 0.05^{a}
Phosphate solubilization at 28 ºC (µg ml^{-1} ± SD)				**pH change Phosphate solubilization at 28 ºC (pH ± SD)**		
isolates	**24hr**	**48hr**	**72hr**	**24hr**	**48hr**	**72hr**
YB1	30.25 ± 2.18^{a}	56.87 ± 0.95^{a}	98.5 ± 0.88^{a}	5.2 ± 0.17^{a}	4.3 ± 0.12^{b}	3.94 ± 0.09^{b}
YB2	32.5 ± 1.95^{a}	60 ± 0.87^{a}	96.87 ± 2.6^{a}	5.39 ± 0.02^{a}	4.63 ± 0.13^{b}	4.06 ± 0.05^{b}
YB3	18.53 ± 1.81^{b}	27.37 ± 2.3^{b}	42.67 ± 1.07^{b}	5.49 ± 0.09^{a}	5.23 ± 0.09^{a}	5.09 ± 0.04^{a}
YB4	27.66 ± 0.91^{a}	55.25 ± 1.52^{a}	98.52 ± 1.2^{a}	5.33 ± 0.13^{a}	4.21 ± 0.19^{b}	3.96 ± 0.09^{b}
YB5	17.55 ± 1.46^{b}	29 ± 1.95^{b}	41.88 ± 1.18^{b}	5.59 ± 0.09^{a}	5.3 ± 0.12^{a}	5 ± 0.19^{a}

Results are means ± SD of three analysis. Means with different superscripts significantly different from each other (P< 0.01)

medium as compared to liquid medium. Strain YB4 efficiently showed P- solubilizing activity in liquid medium. The results were supported by Chaiharn and Lumyong (2011), where some bacterial isolates did not show direct correlation between P-solubilization in solid and liquid medium. Nitrogen fixing ability was shown by strainYB2, YB3 and YB5, in nitrogen free Malate media, by producing a blue colored zone around the colony. Starch hydrolysis was shown by strainYB2 and YB4. Catalase activity was shown by all the selected bacterial isolates except strainYB4, similarly cellulase activity was absent in bacterial strain YB1 (Table 3). These attributes may involve in better growth promotion of plants (Selvakumar *et al*., 2009).

Result showed a significant increase in plant height of Swarna and Swarna sub1 by the inoculation of *Pseudomonas koreensis* as compared to uninoculated rice plants. *Arthrobacter nitroguajacolicus* strainYB4 and strainYB5 significantly increased root length of Swarna sub1. Shoot and root dry weights were significantly increased by all the selected bacterial inoculants except *Arthrobacter nitroguajacolicus* strainYB4 as compared to control plants. The selected isolates increased the phosphorus uptake in both the varieties of rice. In Swarna, strain YB2, YB3, and YB4 whereas in Swarna sub1 all strain except YB3, significantly increased the phosphorus uptake as compared to uninoculated control (Table 4). *In vitro* study clearly revealed that all selected isolates produced IAA, which was responsible for induction and proliferation of lateral roots and root hair in plants (Aloni *et al*., 2006) and solubilization of the inorganic phosphate. Therefore, it may be possible that these bacteria enhanced the rice growth and phosphorus uptake by their attribute of producing IAA and solubilizing phosphate from soil. The enhanced plant growth promotion activities of beneficial microflora have also been reported by Pandey *et al*. (1998); Pandey *et al*. (1999) and Trivedi and Pandey (2007).

In the present investigation, among the selected five bacterial isolates, three were identified as *Arthrobacter* species. The members of genus *Arthrobacter* are prevalent in the environment due to their ability to survive long periods under stressed conditions. They are metabolically diverse and highly resistant to desiccation, starvation and other stresses (Mongodin *et al*., 2006). In this respect, *Arthrobacter nitroguajacolicus* strain YB4, along with *Pseudomonas koreensis* might be of immense considerations for the growth promotion of unique crops growing in the region of diverse climatic conditions of the Indian Himalaya, where temperature fluctuation is frequent and soil faces slight stress due to low level of moisture, carbon and phosphorus availability. These selected bacterial isolates require the development of carrier based inoculants for evaluation and validation of their plant growth promoting activity under field

Table 3 : PGP and biochemical properties of selected bacterial isolates

Strains	Nitrogen activity	Catalase activity	Starch hydrolysis	Cellulose hydrolysis
YB1	-	+	-	-
YB2	+	+	+	+
YB3	+	+	-	+
YB4	-	-	+	+
YB5	+	+	-	+

(-) negative for absence (+) positive for presence

Table 4 : Plant growth promotion by selected bacterial isolates

		Length (cm)		Dry weight (mg pot^{-1})		
Rice variety	Treatments	Shoot	Root	Shoot	Root	P uptake (%)
	Control	19.6 ± 0.70^{a}	11.57 ± 0.87^{a}	53.33 ± 9.39^{a}	23.17 ± 5.35^{a}	0.05 ± 0.006^{a}
	YB1	21.57 ± 1.22^{ab}	12.57 ± 1.70^{a}	164 ± 14.42^{c}	104.5 ± 17.20^{c}	0.119 ± 0.006^{ab}
Swarna	YB2	19.23 ± 1.23^{a}	11.77 ± 1.47^{a}	132.5 ± 16.39^{bc}	80 ± 20^{bc}	0.235 ± 0.054^{c}
	YB3	19.27 ± 0.91^{a}	12.4 ± 1.28^{a}	128.83 ± 21.77^{bc}	82.67 ± 14.29^{bc}	0.142 ± 0.009^{b}
	YB4	25.2 ± 1.30^{b}	11.87 ± 1.59^{a}	90 ± 17.32^{ab}	46.67 ± 5.77^{ab}	0.275 ± 0.014^{c}
	YB5	19.9 ± 0.89^{a}	14.6 ± 1.51^{a}	143.17 ± 15^{bc}	112.5 ± 6.61^{c}	0.098 ± 0.004^{ab}
	Control	19 ± 1.45^{a}	11.03 ± 1.21^{a}	57.83 ± 2.08^{a}	26.83 ± 4.62^{a}	0.046 ± 0.019^{a}
	YB1	23.4 ± 0.79^{b}	11.67 ± 0.95^{ab}	143.33 ± 15.28^{bc}	60 ± 10^{ab}	0.208 ± 0.044^{b}
Swarna sub1	YB2	18.93 ± 1.46^{a}	10.53 ± 0.71^{a}	126.67 ± 20.82^{bc}	58.67 ± 19.19^{ab}	0.202 ± 0.035^{b}
	YB3	22.6 ± 0.85^{ab}	16.03 ± 1.99^{b}	169.83 ± 20.63^{c}	96.33 ± 14.27^{b}	0.111 ± 0.022^{a}
	YB4	19.8 ± 0.56^{ab}	13.4 ± 1.01^{ab}	105 ± 15.17^{ab}	57.67 ± 15.61^{ab}	0.201 ± 0.013^{b}
	YB5	22.93 ± 1.65^{ab}	15.7 ± 1.51^{b}	103.33 ± 15.28^{ab}	60 ± 10^{ab}	0.345 ± 0.056^{c}

Results are means ± SD of three analysis. Mean with different superscript significantly different from each other ($P < 0.01$)

condition in the Indian Himalayan region.

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