BIOTROPIA Vol. 23 No. 2, 2016: 116 - 123 DOI: 10.11598/btb.2016.23.2.511

POTENCY OF RHIZOSPHERE BACTERIA TO PROMOTE RICE GROWTH UNDER SALINE CONDITION

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Received: 8 July 2015/Accepted: 25 July 2016

ABSTRACT

Saline soil is a common problem in coastal paddy field, especially in Indonesia. Salinity affects rice growth and the activities of soil functional microbes, including functional bacteria, which play roles in plant growth. Some of these microbes are associated with rice plants and are able to survive under saline condition. The presence of functional microbes is also important to improve soil quality. Nitrogen and phosphate are essential soil nutrients and is available in soil due to the activities of nitrogen-fixing bacteria and free-living plant-associated bacteria. The objective of the present study was to obtain nitrogen-fixing, phosphate solubilizing and Indole Acetic Acid (IAA)-producing bacteria that are able to survive and promote the growth of rice under saline conditions. From rice and peanut rhizosphere, Ca-phosphate (Ca-P) solubilizing and nitrogen-fixing bacteria were isolated separately using specific media. Then, the Ca-P solubilizing ability, phosphomonoesterase activity and IAA-producing ability were quantitatively examined. Based on the abilities, 20 strains were selected and identified as *Burkholderia cepacia*-complex, *Burkholderia anthina, Burkholderia cenocepacia, Bacillus cereus*-complex (three strains), *Achromobacter spanius, Azospirillum* sp. (four strains), *Azotobacter* sp. (three strains), *Rhizobium leguminosarum*, *Rhizobium* sp. (two strains), and *Pseudomonas* sp. (three strains). The inoculation of several single strains or the mixture of the selected strains promoted the growth of rice under saline conditions. These inoculants could be potential as biofertilizer in saline paddy fields.

Keywords: Indole Acetic Acid production, phosphate solubilization, plant growth promoting bacteria, nitrogen fixation, rhizosphere, rice

INTRODUCTION

Most of the fertile paddy fields in Indonesia are located in coastal area and experiences soil salinization due to seawater intrusion (Djufry *et al.* 2011). Salinity affects not only the growth of rice (*Oryza sativa* Linn.), but also the activities of functional soil microbes, including bacteria, that play roles in mineralization of macro and microelements for plant growth (Balser *et al.* 2006). Some of these bacteria are associated with rice plants and are able to survive under saline condition. The activity of soil microbes is an important aspect of biogeochemical cycles of carbon, nitrogen, sulfur, phosphorus, etc. (Banig *et al.* 2008). The presence of functional microbes is also important to improve the quality of soil (Wijebandara *et al.* 2009). Nitrogen and phosphate are essential nutrients and are available in soil due to the activities of nitrogen-fixing bacteria and free-living plant-associated bacteria (Steenhoudt & Vanderleyden 2000). Several bacteria belonging to the genera *Rhizobium*, *Azotobacter* and *Azospirillum* are able to fix nitrogen and solubilize phosphate (Nosrati *et al.* 2014). Some members of these genera also produce plant growth promoting hormone such as Indole Acetic Acid (IAA), gibberellins and cytokinins (Bhattacharyya & Jha 2012). Therefore, these genera are regarded as important components of biofertilizer (Rao 1994; Bhattacharjee & Dey 2014). Introduction of growth promoting bacteria can increase nitrogen availability for plants and enhance crop productivity. However, very little information is available for the effect of salinity on bacteria that have beneficial functions, such as nitrogen fixation, phosphate solubilization and the production of plant growth hormone (Pliego *et al.* 2011; Lugtenberg *et al.* 2013; Nakbanpote *et al.* 2014). The purpose of this study is to obtain nitrogen-fixing, phosphate solubilizing and IAA producing bacteria that are able to survive and

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promote the growth of rice under saline conditions.

MATERIALS AND METHODS

Bacterial Sources

Bacterial sources were obtained from collected rhizosphere of rice (*Oryza sativa*) and peanut (*Arachis hypogaea*) cultivated in the research field of Cibinong Science Center, West Java Province, Indonesia. The physical and chemical properties of this research field indicated that the soil is infertile soil (Table 1).

Isolation of Bacteria

Phosphate solubilizing bacteria were screened following the method of Park *et al.* (2011). Halo zone formation around colonies after 7 day-cultivation at 30 °C on Pikovskaya medium was used as an indicator of Ca-Phosphate (Ca-P) solubilization (Nguyen *et al.* 1992). Nitrogen-fixing bacteria were isolated targeting the genera *Rhizobium, Azopirillum* and *Azotobacter* according to Mubarik *et al.* (2011) and Salamone *et al.* (2012) as well as Aquilanti and Clementi (2004), respectively.

Determination of Ca-P Solubilization

Ca-P solubilizing ability of the isolated strains was quantitatively determined according to Chen *et al.* (2006) by measuring orthophosphate in the culture fluid after 7 days of cultivation. Orthosphosphate determination was conducted according to Vassileva *et al.* (2000).

Determination of Phosphatase Activity

Extracellular phosphomonoesterase (PMEase) activity of the strains was determined following the method of Tabatabai and Bremner (1969) using p-nitrophenyl phosphate. The unit of the PMEase activity was defined as μmol/hof p-nitrophenol released in 1 mL of extracellular enzyme solution that was fractioned from 1.0 mL of the culture fluid after 7 days of cultivation.

Determination of IAA Production

The IAA production of the strains was investigated after 7 days of cultivation, following the methods of Crozier *et al.* (1988) and Gravel *et al.* (2007).

Selection and Identification of Bacteria

Based on the Ca-P solubilization, PMEase activity and IAA production abilities, a total of 20 strains were selected from the above strains for rice growth assays. Identification of the 20 strains was performed following the method of Otsuka *et al.* (2008) based on the 16S rRNA gene sequence with 16S-9F (5-GAGTTTGATCCTGGCTCAG-3) and 16S-1510R (5-GGCTACCTTGTTACGA-3) primers.

Rice Growth Assay at the Stage of Germination under Saline Condition

One strain out of 10 taxonomic groups was selected and subjected to a root and shoot growth assay of rice at the stage of germination based on Zaller (2007) and Cerabolini *et al.* (2004). Briefly, ten seeds of three rice cultivars, INPARA-3, INPARI-13 and INPARA-6 were soaked in sterile water for 5 hours in their respective containers. These 10 rice seeds were then arranged based on their respective cultivars on top of filter paper which was put inside a Petridish (20 cm in diameter). Fifteen milliliter of 4.0 g/L NaCl solution was poured onto the filter paper inside

Table 1 Physical and chemical properties of soils in the research field

Parameter	P (%)	K (%)	C (%)	N (%)	C/N ratio	Ca (%)	Exchangable Mg (%)	Exchangable Na (%)	Exchangable Al dd (%)	Soil pH	Amount of bacteria population
Characteristic	0.173	0.045	1.303	0.36	3.61	11.41	0.57	0.30	0.04	5.8	10^4-10^5
Determined accoding to Rowell (1994)	Very low	Very low	Low	Moderate	Very low	High	Low	Low	Low	Acid	Infertile

each Petridish, on which 10 rice seeds were lined up, followed by 1.0 mL of bacterial inoculant suspension containing 10^9 cells/mL. Root and shoot lengths were measured at 7 days after germination. This experiment was set up using Complete Randomized Design with three replications.

Rice Growth Assay at 45 Days after Planting under Saline and Non-Saline Conditions

Ten strains out of 20 isolates tested on germination test were then subjected to rice growth assay for 45 days under saline condition with 0.4% NaCl. The number of cells for each treatment was adjusted to about 3.2 x 10^7. This value was selected based on the number of bacteria commonly found in paddy field soil. In a preliminary test (rice growth assay at the stage of germination), INPARI-13 and INPARA-6 could not grow well under the same saline condition. Therefore, only INPARA-3 was used in this assay. Four seeds of INPARA-3 were planted to experimental pots (0.5 gallon pots) containing sterile sands (1.5 kg) flooded with water(field capacity of sands = 24% or 360 mL). Treatments applied were: 1. Saline condition (adding 360 mL of 0.4% NaCl (6 g NaCl) to the 0.5 gallon pots) and 2. non-saline condition (without 0.4% NaCl). Into each pot, 5 mL of bacterial inoculant suspension was added. The result of experiment is shown in Table 5. After 7 days, the second inoculation with the same amount of bacterial suspension was conducted. The water level in pot was regulated by adding sterile water to compensate water decrease due to evaporation. The electrical conductivity (EC) value of the assay media under saline condition was kept at 7.5 mS/cm. At 45 days after planting the growth of rice was evaluated. This experiment was set as complete randomized design performed with three replications.

RESULTS AND DISCUSSION

Composition of the Strains

The selected 20 strains, originated from the rhizosphere of rice and peanut, belonged to the genera *Burkholderia, Bacillus, Achromobacter, Pseudomonas, Azospirillum, Rhizobium* and *Azotobacter* (Table 2). The selected strains were originated from non-saline soil. The reason for the selection was to compare the physiological characteristics of microbes isolated from saline and non-saline soil. The result of this study showed that the functional microbes for

Table 2 List of bacteria isolated from rice and peanut rhizosphere

Isolate code*	Phylum/class**	Taxon	Source (rhizosphere)
CSC P1	Proteobacteria/Beta-	*Burkholderia cepacia*-complex	Rice
CSC P2	Proteobacteria/Beta-	*Burkholderia cenocepacia*	Rice
CSC P3	Firmicutes/Bacilli	*Bacillus cereus*-complex	Rice
CSC P4	Proteobacteria/Beta-	*Achromobacter spanius*	Rice
CSC P5	Firmicutes/Bacilli	*Bacillus cereus*-complex	Rice
CSC P6	Proteobacteria/Gamma-	*Pseudomonas* sp.	Rice
CSC P7	Proteobacteria/Alpha-	*Azospirillum* sp.	Rice
CSC P8	Proteobacteria/Alpha-	*Azospirillum* sp.	Rice
CSC P9	Proteobacteria/Alpha-	*Rhizobium* sp.	Rice
CSC P10	Proteobacteria/Gamma-	*Azotobacter* sp.	Rice
CSC P11	Proteobacteria/Gamma-	*Azotobacter* sp.	Rice
CSC P12	Proteobacteria/Beta-	*Burkholderia anthina*	Rice
CSC N1	Proteobacteria/Alpha-	*Rhizobium* sp.	Peanut
CSC N2	Proteobacteria/Gamma-	*Pseudomonas* sp.	Peanut
CSC N3	Proteobacteria/Alpha-	*Rhizobium leguminosarum*	Peanut
CSC N7	Proteobacteria/Alpha-	*Azospirillum* sp.	Peanut
CSC N8	Firmicutes/Bacilli	*Bacillus cereus*-complex	Peanut
CSC N9	Proteobacteria/Gamma-	*Azotobacter* sp.	Peanut
CSC N10	Proteobacteria/Gamma-	*Pseudomonas* sp.	Peanut
CSC N11	Proteobacteria/Alpha-	*Azospirillum* sp.	Peanut

Notes: * = A Strain with P in its code were isolated as Ca-P solubilizing bacteria, and that with N were isolated as nitrogen fixing bacteria
** = *Alpha-*, *Beta-* and *Gamma-* denote the classes *Alphaproteobacteria*, *Betaproteobacteria* and Gammaproteobacteria, respectively

promoting rice growth were not different from that reported by Susilowati *et al.* (2015).

Phosphate Solubilizing Ability of the Strains

Ca-P solubilizing ability of the strains is shown in Table 3. The difference in the strength of Ca-P solubilizing ability was not related to the taxonomic property. All strains formed halo zone around colonies, and the area ratio of the halo zone to a colony was variable (data not shown) indicating the ability to solubilize Ca-P differed among the strains. This was reflected in the Ca-P solubilizing ability which was quantitatively determined (Table 3). The highest Ca-P solubilization ability was shown by *Pseudomonas* sp. CSC N2 and the lowest was shown by *Achromobacter spanius* CSC P4. The activity of PMEase is shown in Table 3. *Pseudomonas* sp. CSCN2 again showed the highest PMEase activity, and *Achromobacter spanius* CSC P4 seemed to have no extracellular PMEase activity.

Nitrogen-Fixing Ability of the Strains

All strains belonging to *Rhizobium*, *Azotobacter* and *Azospirillum* genera were able to grow on nitrogen-limited media implying that these strains were able to fix nitrogen (Chien *et al.* 1992).

IAA Production of the Strains

IAA production of the strains is shown in Table 3. The amount of IAA produced varied depending on strains. The highest production was achieved by *Azospirillum* sp. CSC P8 and *Azospirillum* sp. CSC P7. The lowest IAA production was detected in *Achromobacter spanius* CSC P4.

Effect of Bacterial Inoculation on the Rice

During the 7-day germination assay with 0.4% NaCl, the effect of bacterial inoculation varied depending on the strains (Table 4). The best growth was obtained by the mixture of strains on INPARA-3, with 7.46 cm and 6.5 cm in shoot and root length, respectively. Medium level effect was observed in *Burkholderia cepacia*-complex, *Bacillus cereus*-complex, *Pseudomonas* sp., *Azospirillum* sp. and *Azotobacter* sp. However, inoculation of *Burkholderia cenocepacia, Achromobacter spanius, Rhizobium* sp., *Burkholderia anthina* and *Rhizobium leguminosarum* had no effect on shoot and root length. Cultivars INPARA-6 and INPARI-13 could not grow without any inoculant (control) or with five single-strain-inoculants.

In the 45-day growth assay (Table 5), the growth of rice cultivar INPARA-3 under saline

Table 3 $Ca_3(PO_4)_2$ solubilization ability, PMEase activity and IAA production of the strains

Isolate code	Taxon	Phosphate Solubilization (mg/L)*	PMEase (Unit)*	IAA Production (mg/L)*
CSC P1	*Burkholderia cepacia*-complex	8.72 ± 0.89	0.63 ± 0.71	8.67 ± 0.92
CSC P2	*Burkholderia cenocepacia*	1.06 ± 0.16	0.13 ± 0.52	2.63 ± 0.16
CSC P3	*Bacillus cereus*-complex	10.54 ± 0.16	0.82 ± 0.85	8.16 ± 0.90
CSC P4	*Achromobacter spanius*	0.30 ± 0.68	0.01 ± 0.04	1.94 ± 0.21
CSC P5	*Bacillus cereus*-complex	1.51 ± 0.11	0.10 ± 0.86	5.46 ± 0.58
CSC P6	*Pseudomonas* sp.	11.26 ± 0.58	0.75 ± 0.26	8.27 ± 0.67
CSC P7	*Azospirillum* sp.	7.39 ± 0.42	0.60 ± 0.86	9.45 ± 0.06
CSC P8	*Azospirillum* sp.	6.68 ± 0.37	0.68 ± 0.10	9.56 ± 0.16
CSC P9	*Rhizobium* sp.	2.28 ± 0.63	0.51 ± 0.70	6.08 ± 0.42
CSC P10	*Azotobacter* sp.	5.71 ± 0.53	1.27 ± 0.68	8.75 ± 0.98
CSC P11	*Azotobacter* sp.	1.57 ± 0.95	0.49 ± 0.67	6.21 ± 0.32
CSC P12	*Burkholderia anthina*	0.47 ± 0.47	0.10 ± 0.12	2.13 ± 0.16
CSC N1	*Rhizobium* sp.	1.18 ± 0.05	2.01 ± 0.34	3.82 ± 0.89
CSC N2	*Pseudomonas* sp.	11.39 ± 0.53	2.22 ± 0.93	8.16 ± 0.90
CSC N3	*Rhizobium leguminosarum*	4.94 ± 0.32	0.31 ± 0.27	8.61 ± 0.10
CSC N7	*Azospirillum* sp.	2.00 ± 0.32	0.47 ± 0.03	7.61 ± 0.39
CSC N8	*Bacillus cereus*-complex	0.89 ± 0.95	0.12 ± 0.88	2.73 ± 0.68
CSC N9	*Azotobacter* sp.	0.83 ± 0.89	0.45 ± 0.09	8.39 ± 0.06
CSCN10	*Pseudomonas* sp.	10.08 ± 0.26	0.85 ± 0.89	8.33 ± 0.84
CSCN11	*Azospirillum* sp.	1.86 ± 0.47	0.14 ± 0.59	8.09 ± 0.22

Note: Values represent mean±standard deviation (n = 3)

Table 4 The effect of bacterial inoculants on root and shoot length of rice (three cultivars) at the stage of seed germination

Isolate code	Taxon	Rice cultivar	Shoot length (cm)*	Root length (cm)*
Control	(Control: no inoculation)	INPARA-3	4.03 a	0.51 a
		INPARI-13	dead	dead
		INPARA-6	dead	dead
CSC P1	*Burkholderia cepacia*-complex	INPARA-3	5.55 de	4.43 ghi
		INPARI-13	4.23 ab	2,00 bcde
		INPARA-6	4.37 ab	1.39 abc
CSC P2	*Burkholderia cenocepacia*	INPARA-3	4.16 ab	1.47 abcd
		INPARI-13	dead	dead
		INPARA-6	dead	dead
CSC P3	*Bacillus cereus*-complex	INPARA-3	5.54 de	4.24 ghi
		INPARI-13	4.51 abcd	2.99 defg
		INPARA-6	4.59 abcd	2.04 bcde
CSC P4	*Achromobacter spanius*	INPARA-3	4.09 a	2.90 cdefg
		INPARI-13	dead	dead
		INPARA-6	dead	dead
CSC P6	*Pseudomonas* sp.	INPARA-3	5.58 de	4.70 hi
		INPARI-13	4.55 abcd	3.41 efgh
		INPARA-6	4.79 abcd	3.22 efgh
CSC P8	*Azospirillum* sp.	INPARA-3	6.10 e	5.00 i
		INPARI-13	4.80 abcd	3.65 fghi
		INPARA-6	4.94 abcd	3.11 efgh
CSC N1	*Rhizobium* sp.	INPARA-3	4.30 ab	3.95 fghi
		INPARI-13	dead	dead
		INPARA-6	dead	dead
CSC N9	*Azotobacter* sp.	INPARA-3	5.56 de	4.44 ghi
		INPARI-13	4.82 abcd	2.83 cdefg
		INPARA-6	4.31 ab	2.39 bcdef
CSC N12	*Burkholderia anthina*	INPARA-3	4.03 a	1.25 ab
		INPARI-13	dead	dead
		INPARA-6	dead	dead
CSC N3	*Rhizobium leguminosarum*	INPARA-3	4.34	3.62
		INPARI-13	dead	dead
		INPARA-6	dead	dead
Mix	Mixture of strain	INPARA-3	7.46 f	6.50 j
		INPARI-13	4.98 abcd	4.01 ghi
		INPARA-6	4.96 abcd	4.13 ghi

Note: Values followed by the same letter in the same column are not significantly different based on Duncan's multiple range test at 5% level

condition was less than that under no saline condition. Under saline condition, rice cultivar INPARA-3 inoculated with the mixture of strains showed the best growth with 29 cm in plant height and 5.5 cm in root length. As a single isolate inoculation, *Pseudomonas* sp. CSC N6 showed the best effect.

Twenty strains with Ca-P solubilizing, extracellular PMEase producing and IAA producing abilities were successfully obtained, with an exception of *A. spanius* CSC P4 that did not show clear PMEase activity. These strains did not lean to a specific taxonomic lineage and composed of members of the phyla *Proteobacteria* (the classes *Alphaproteobacteria*, *Betaproteobacteria* and *Gammaproteobacteria*) and *Fermicutes*. The fact that the strains were isolated as nitrogen-fixing bacteria including Ca-P solubilizing members indicated that Ca-P solubilizing ability was common among bacteria, at least among those living in the rhizosphere. It was also possible that PMEase activity was common among

Table 5 The effect of bacterial inoculants on the growth of rice cultivar INPARA-3, in sterile sand media under saline and non-saline conditions 45 days after planting

Isolate code	Inoculant	Salinity condition	Total dry biomass (g)	Plant height (cm)	Root length (cm)
–	No bacteria	Non-saline	0.02 a	14.25 ab	1.50 ab
		Saline	0.01 a	13.00 a	1.00 a
CSC P1	*Burkholderia cepacia*-complex	Non-saline	0.09 cde	27.50 ghi	5.00 jk
		Saline	0.07 abcd	26.25 ghi	4.00 hij
CSC P2	*Burkholderia cenocepacia*	Non-saline	0.06 abcd	26.00 fgh	3.50 fgh
		Saline	0.04 abc	22.75 cdefg	2.65 de
CSC P3	*Bacillus cereus*-complex	Non-saline	0.09 cde	27.30 ghi	4.50 ij
		Saline	0.08 bcd	26.00 fgh	3.75 ghi
CSCP4	*Achromobacter spanius*	Non-saline	0.04 abc	25.50 efgh	3.50 fgh
		Saline	0.02 a	17.00 abc	3.00 ef
CSC P6	*Pseudomonas* sp.	Non-saline	0.13 e	29.50 hi	6.15 l
		Saline	0.09 cde	26.88 ghi	5.00 jk
CSC P8	*Azospirillum* sp.	Non-saline	0.09 cde	27.50 ghi	4.25 ij
		Saline	0.07 abcd	26.25 ghi	4.00 hij
CSC N1	*Rhizobium* sp.	Non-saline	0.07 abcd	24.50 defgh	3.25 fg
		Saline	0.04 abc	20.75 cde	2.25 cd
CSC N9	*Azotobacter* sp.	Non-saline	0.08 bcd	26.50 ghi	4.50 ij
		Saline	0.07 abcd	26.25 ghi	3.75 ghi
CSCN12	*Burkholderia anthina*	Non-saline	0.06 abc	21.00 cdef	3.50 fgh
		Saline	0.02 a	17.15 abc	2.25 cd
CSC N3	*Rhizobium leguminosarum*	Non-saline	0.05 abc	23.00 defg	3.25 fg
		Saline	0.02 abc	19.00 bcd	2.00 bc
–	Mixture of all the isolates	Non-saline	0.14 e	31.00 i	6.50 l
		Saline	0.12 de	29.00 hi	5.50 k

Notes: Values followed by the same letter in the same column are not significantly different by Duncan's multiple range test at 5% level.
Non-saline condition = 360 mL freshwater in 0.5 gallon pots.
Saline condition = 360 mL freshwater in 0.5 gallon pots was added with 0.4% NaCl (6 g NaCl).

rhizosphere bacteria. Interestingly, the strains with higher Ca-P solubilizing ability generally showed higher PMEase activity. IAA production was also reported as common among soil bacteria (Hasan 2002; Xin *et al.* 2009), which was supported by the present study.

Saline environment inhibits rice growth. This is because rice is a saline sensitive plant (Ashraf & Harris 2004); also because the uptake of Ca_2^+, K^+ and inorganic N and P are disrupted under high Na concentration (Ashraf & Harris 2004). In addition, the salinity also affected soil enzyme activities (Siddikee *et al.* 2011), which could indirectly affect rice growth. The inoculation of the selected strains affected germination of rice under saline condition (Table 4). The inoculation of mixture of the strains resulted in the best rice growth. As single strain, *Azospirillum* sp. CSC P8 and *Pseudomonas* sp. CSC P6 provided the best and the second best rice growth support, respectively. It was possible that the inoculants supported the growth of rice by supplying phosphate and IAA. *Azospirillum* sp. is a potential nitrogen fixer and the mixture of the strains also includes nitrogen fixers. Therefore, it was possible that nitrogen fixed by the inoculants might also promote rice growth. Rice cultivars INPARI-13 and INPARA-6 did not grow without the existence of inoculants. The present study showed that the inoculation of five strains and the mixture of strains enabled these cultivars to grow. This indicated that the inoculation not only promoted rice growth by supplying nutrient and IAA, but also enhanced rice tolerance towards salinity. It was interesting that some strains isolated from peanut rhizosphere could promote and support rice growth.

Among the rice cultivars tested in the present study, only INPARA-3 grew in saline condition without inoculation of the strains. Therefore, INPARA-3 was then subjected to rice growth assay with 0.4% NaCl. In this assay, the inoculation of the mixture of strains, *Pseudomonas* sp. CSC P6 and *Azospirillum* sp. CSC P8 provided

the best, the second best, and the third best rice growth support, respectively. These inoculants may be promising as biofertilizer to support rice growth in saline paddy fields.

CONCLUSIONS

Twenty strains of rhizosphere bacteria with Ca-P solubilizing ability and IAA production were successfully obtained in this study. Those bacteria mainly belonged to *Burkholderia cepacia*-complex, *Burkholderia anthina, Burkholderia cenocepacia, Bacillus cereus*-complex, *Achromobacter spanius, Azospirillum* sp., *Azotobacter* sp., *Rhizobium leguminosarum, Rhizobium* sp. and *Pseudomonas* sp.

Potential nitrogen fixing bacteria are *Azospirillum* sp., *Azotobacter* sp., *Rhizobium leguminosarum* and *Rhizobium* sp. Most strains had PMEase activity. Some strains showed growth-promoting effect on rice under saline conditions and produced plant growth hormone. These strains could be candidates for biofertilizer for rice in saline paddy field. It is also important to consider using combination of inoculants and rice cultivars to obtain maximum result.

ACKNOWLEDGEMENTS

This work was funded by JICA-JST SATREP 2011-2016. We thank Dr Shigeto Otsuka from University of Tokyo for research guidance. We are grateful to Senlie Oktavitana, Anis Mutirani and Rinatu Siwi for their assistance in laboratory works.

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