

Research Article

Isolation and Characterization of Phosphate Solubilizing Bacteria from Upland Rice Cultivation Areas in Bangka Regency

Kartika Kartika^{1,2}, Abdul Munif³, Endah Retno Palupi⁴, Satriyas Ilyas⁴, Mohamad Rahmad Suhartanto⁴*

1)Postgraduate doctoral student of Seed science and Technology Study Program, Department of Agronomy and Horticulture, IPB University IPB University, Jl. Meranti, Babakan, Kec. Dramaga, Kabupaten Bogor, Jawa Barat 16680

2)Bangka Belitung University Kampus Terpadu Universitas Bangka Belitung Desa Balunijuk, Kecamatan Merawang, 33172
3)Phytopathology Study Program, Department of Plant Protection, IPB University, Jl. Meranti, Babakan, Kec. Dramaga, Kabupaten Bogor, Jawa Barat 16680

4)Seed science and Technology Study Program, Department of Agronomy and Horticulture, IPB University, Jl. Meranti, Babakan, Kec. Dramaga, Kabupaten Bogor, Jawa Barat 16680

* Corresponding author, email: m.r.suhartanto@apps.ipb.ac.id

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ABSTRACT

The availability of phosphorus (P) in ultisol acid soils presents a significant challenge due to its attachment to aluminum (Al) or iron (Fe) compounds. A potential solution to address this issue is the utilization of phosphate solubilizing bacteria (PSB). Therefore, this study aimed to analyze the potential of PSB originating from upland rice cultivation on ultisol soils. The bacterial isolates were obtained from soil samples taken from the rhizosphere area and root tissue of upland rice plants cultivated in Payabenua and Saing Villages, Bangka Regency. The pathogenicity testing encompassed hypersensitivity and hemolysis tests, while the P solubilization included the evaluation of the phosphate solubilizing index (PSI) and P dissolution. Subsequently, the selected isolates were subjected to phosphatase enzyme and organic acid content assessment. The results showed a total of 120 isolates, predominantly distributed in the Payabenua area and primarily consisting of endophytic bacteria. Among the six selected isolates, genus Burkholderia dominated four isolates, while the remaining isolates belonged to genus Serratia. Furthermore, in Burkholderia vietnamiensis, the solubility value of P in AlPO4 and Ca3(PO4)2 liquid media exhibited a range of 0.0013 to 0.0344% and 0.0008 to 0.1842%, respectively.

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INTRODUCTION

Farmers in Bangka Belitung Province have a tradition of cultivating padi gogo, which is locally known as "berhume". This cultivation activity is typically conducted in upland areas that are newly opened or in between black pepper and rubber plantations. Among the districts in the province, Bangka Regency serves as a central location for padi gogo cultivation. However, the soil has a pH below 5 and is classified as an ultisol soil type. Wahyudin et al. (2017) and Asril and Lisafitri (2020) highlighted that ultisol soils typically exhibited low nutrient content. The essential nutrient phosphorus (P) is bound to aluminum (Al) and iron (Fe), rendering it in an insoluble phosphate form. Consequently, a significant portion of the P is not readily available for plant absorption and utilization.

Phosphate solubilizing bacteria (PSB) offers a viable approach for releasing P element form of Al and Fe bonds within ultisol soil. By harnessing the metabolic abilities, the bonds between Al and Fe in the ultisol soil can be effectively released, thereby facilitating the availability of phosphates for plant uptake (Sugianto et al. 2019; Sonia & Setiawati 2022). Numerous studies have been undertaken to investigate the efficacy of bacterial inoculation in enhancing P availability. Setiawati and Pranoto (2015) highlighted that the capacity of these bacteria to solubilize phosphate faced limitations when introduced into different environments. Furthermore, Awais et al. (2017) emphasized the variability in population sizes of PSB across different soil types.

PSB is able to convert insoluble phosphates into forms available to plants through the secretion of organic acids (Pande et al. 2020) and the production is one of the indicators of the activity of PSB (Fitriatin et al. 2020). The organic acids produced by PSB are chelating agents for calcium (Ca), magnesium (Mg), Fe, and Al to form stable complexes (Yadav et al. 2015). Meanwhile, the amount and type of organic acids vary between microorganisms (Krishnaraj & Dahale 2014).

Isolation of PSB from rice crops has been carried out by (Putriani et al. 2019; Hartanti 2020; Wiraswati et al. 2020; Damo et al. 2022). *Enterobacter asburiae* is PSB isolated from the planting of paddy in the Aceh region (Putriani et al. 2019). Genus *Pseudomonas*, genus *Bacillus*, genus *Enterobacter*, and genus *Azotobacter* are PSB that are isolated from the paddy plant Situbagendit (Hartanti 2020). *Basil* sp., *Enterobacter* sp., and *Brachybacterium* sp. bacteria are also isolated from the paddy plant philosopher originating in West Java (Wiraswati et al. 2020). Furthermore, *Acidovorax* sp., *Pseudomonas* sp., *Burkholderia* sp., *Sphingomonas* sp., *Mycolicibacterium* sp., and *Variovorax* sp. are PSB isolated from paddy field soils in Japan (Damo et al. 2022).

PSB isolated from the cultivated land of paddy has several capabilities as Indole Acetic Acid (IAA) producers, inhibitors of pathogens, and biological control agents. Enterobacter asburiae is also a PSB isolated from the Aceh region with the ability to produce IAA hormones (Putriani et al. 2019). According to Parida et al. (2017), Bacillus subtilis shows promising potential as an inducer of resistance against HDB (Hawar Daun Bakteri/Bacterial Leaf Blight) disease in paddy plants. Furthermore, Wiraswati et al. (2020) highlighted its effectiveness as a biological control agent, owing to the production of an anti-fungal compound that aids in combatting Blast disease. Munif and Nurjayadi (2021) also identified several endophytic bacteria isolates, namely GH1, Si2, Si33, Sp24, and G053, which possess the capability to control Meloidogyne graminicola in rice crops. In another study conducted by Prihatiningsih et al. (2021), three endophytic bacteria strains are identified as potential agents for promoting plant growth and controlling bacterial leaf blight on rice.

Klebsiella and Acinetobacter are two isolates of PSB isolated from Pekanbaru's ultisol soil (Oksana et al. 2020). There is a scarcity of study focused on microbes, particularly PSB, originating from the ultisol soil of Bangka Island. According to Prihastuti (2012), several isolates isolated from ultisols are useful isolates. In addition, the original isolate originating from an area has adaptability to the local environment so that it is easier to apply it again to that area. Therefore, this study conducts an inventory and assessment of the potential PSB derived from paddy gogo cultivation in Bangka's ultisol.

MATERIALS AND METHODS Materials

Soil samples were collected from upland rice cultivation areas located in Payabenua and Saing regions of Mendo Barat District, Bangka Regency. The samples were processed using pikovskaya medium (5 g of AlPO₄; 0.5 g (NH₄)₂SO₄, 0.1 g MgSO₄.2H₂O, 0.001 g MnSO₄, 0.001 g FeSO₄, 0.5 g yeast extract, and 15 g agar), 2% NaOCl, 70% alcohol, the tobacco leaf, sterile aquadest, Tryptone Soya Agar (TSA) media, Blood Agar media, 1% glucose, 0.05% yeast extract, and 0.5% phosphate, boric acid 0.5%, ammonium molybdate 0.38%, HCl 7.5%, KH₂PO₄, universal primers (Primer F: 16F27 and Primer R: 16R 1492).

Methods

The followings are four steps in this study.

Isolation of Rhizosphere and Endophytic Bacteria

Soil sampling was conducted during both the vegetative and generative phases of the study. The bacteria isolated were identified as rhizospheric and endophytic bacteria. The rhizospheric bacteria were derived from the soil surrounding the roots of the upland rice plant. To begin the process, 10 g soil samples were collected and subsequently dissolved in 90 ml of sterile aquadest. The resulting solution was then shaken vigorously for 2 minutes, resulting in a dilution of 10⁻¹. Subsequently, 1 ml of the soil solution was transferred to a test tube containing 9 ml of sterile aquadest, and the mixture was agitated using a vortex to achieve a dilution of 10⁻². This process was repeated until a dilution of 10⁻⁷ was obtained. From each series, 1 ml was aseptically transferred to a petri dish containing Pikovskaya agar medium. Petri dishes containing bacterial inoculations were incubated at room temperature for 3-6 days till a clear zone appeared. Isolates with clear zones were taken aseptically with a sterile needle and scratched on the agar medium as selected isolates.

Endophytic bacteria were isolated from the roots of the upland rice plant and carefully washed under a continuous stream of running water. The washed roots were then cut into 1-2 cm fragments and soaked in running water for 1-2 hours. Subsequently, the roots were dried on sterile tissue and to ensure the sterility of the surface, a sterilization procedure was conducted. The surface of the roots was treated with 2% NaOCl for 2-3 minutes, followed by a 70% alcohol treatment for 1-2 minutes. To eliminate any residual contaminants, the roots were then rinsed with sterile aquadest, repeating the rinsing process up to three times for 1 minute each time. Finally, they were dried again on sterile tissue and the browning areas were carefully removed, ensuring the integrity of the samples. The roots were then weighed 1 g and crushed using a sterile mortar. To obtain a root extract suitable for further analysis, the crushed roots were mixed with 9 mL of sterile aquadest and diluted accordingly to achieve a ratio of 10-4. For the cultivation of the bacterial suspensions, 100 µL of the diluted root extract was inoculated onto Tryptone Soya Agar (TSA) media using surface plating method. The inoculated media plates were then incubated at room temperature for 48 hours, allowing the growth and development of the bacterial colonies.

Pathogenicity Testing

This study conducted pathogenicity testing, which comprised hypersensitivity and hemolysis tests. The hypersensitivity test method employed in this study was developed by Ropalia (2015). Bacterial colonies approximately 5 mm in diameter were collected using a loop and subsequently suspended in 2.5 mL sterile water. The amount of 0.5 mL suspension was then injected onto the lower surface of the leaf, without penetrating the upper layer. After inoculation, the leaves were incubated for 24-48 hours, during which observations were made to detect any symptoms on the tobacco leaves. A positive hypersensitive reaction was identified by the presence of necrotic symptoms on the injected tobacco leaves. However, isolates that exhibited negative reactions were classified as nonpathogenic. The hemolysis test involved growing bacteria selected during the hypersensitivity test on Blood Agar medium, followed by an incubation period of 24 hours at room temperature. The presence of a clear zone surrounding the perimeter of the isolate indicated pathogenicity to both humans and animals.

Testing of the isolates in phosphate solubilization

The test of PSB ability to dissolve phosphates consisted of the phosphate solubilizing index (PSI) and the P dissolution test. P nutrient solubilizing index test was carried out by streaking the PSB isolates on tricalcium phosphate agar medium, which was a modification of Pikovskaya media consisting of 10 g glucose, 5 g $Ca_3(PO_4)_2$, 0.5 g $(NH_4)2SO_4$, 0.2 g KCL, 0.1 g MgSO₄.7H₂O, 0.5 g yeast extract, 25 mg MnSO₄, and 25 mg FeSO₄ as well as 20 g agar in 1 L aquadest. Furthermore, isolates of bacteria aged 48 hours were taken using sterile oasis needles and grown on Pikovskaya medium. The clear zone around isolates was observed 7 days after incubation and the PSI was measured using the following formula:

 $PSI = \frac{clear \text{ zone diameter - isolate diameter}}{isolate diameter}$

The P dissolution test was carried out by isolating bacteria on 25 ml of the growing medium for 7 days. The growing medium used consists of 1% glucose, 0.05% yeast extract, and 0.5% phosphate binding sources (Al and Ca). After incubation, the suspension of isolates was subjected to centrifugation at 4000 rpm for 25 minutes. This process aimed to separate the supernatant from microbial cells and insoluble phosphates. Following centrifugation, the supernatant was carefully collected and subsequently filtered. The measurement of dissolved phosphate was carried out by using modified method of Susilowati and Syekhfani (2014), in which 1 ml of the supernatant is mixed with reagent solution (boric acid 0.5%, ammonium molybdate 0.38%, HCl 7.5%) with 5 drops of reducing solution. The reaction solution was measured using a spectrophotometer with a wavelength of 880 nM and a standard solution using KH₂PO₄. Based on the results of phosphate measurements in liquid media, the percentage of phosphate solubility was calculated to determine the amount of PSB dissolving in phosphates. The percentage of phosphate solubility is calculated by the formula:

Solubility Percentage (%) =
$$\frac{\text{available P level (ppm)}}{\text{Total P level at source (ppm)}} \ge 100\%$$

Testing of Organic Acids Content, Enzyme Phosphatase and Molecular Identification of Selected Bacteria Isolates

Selected PSB isolates were sent to the test laboratory to determine the content of organic acids, phosphatase enzymes, and molecular identification. Extraction of organic acids was carried out in Bogor Agrochemical Residue Laboratory using the HPLC analysis method. Measurement of phosphatase enzyme activity was conducted in the Soil Biology Laboratory, Faculty of Agriculture, Padjajaran University, using a spectrophotometer at a wavelength of 400 nm. Furthermore, the molecular identification of bacterial isolates was conducted in Bogor Environmental Biotechnology Laboratory for sequencing 16 Sr DNA gene with universal primers (Primer F: 16F27 and Primer R: 16R 1492).

RESULTS AND DISCUSSION Result

Isolate of Rhizospheric and Endophytic Bacteria

The number of PSB isolates from the rhizosphere and endophytes of field rice cultivation during the vegetative and generative phases at different locations is shown in Table 1. The total isolates of PSB isolated from rice field cultivation areas at all locations in Bangka Regency were 120 isolates with the most distribution in the Payabenua area with 76 isolates. The 120 bacterial isolates were dominated by endophytic bacteria, namely 109 isolates, and 68 were found in the vegetative phase.

Pathogenicity Testing

About 120 isolates were successfully isolated and subjected to hypersensitivity testing. According to the results detected, 64 bacterial isolates caused chlorosis in tobacco leaves (Table 2) that caused chlorosis in tabacco leaves (Figure 1).



Figure 1. Tobacco leaves showing chlorosis after being tested for hypersensitivity.

Furthermore, 29 out of the 40 PSB isolates tested for hemolysis passed. Bacterial isolates that cause hemolysis on blood agar media are shown in Figure 2.



Figure 2. Clear zones (indicated by arrows) formed in hemolysis test.

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Location	Sample	Growth phase	Rhizospheric bacteria	Endophytes bacteria				
			isolate					
	1	Vegetative	0	14				
Payabenua	1	Generative	4	1				
-	2	Vegetative	5	15				
		Generative	0	37				
	Total		9	67				
	1	Vegetative	0	37				
S	1	Generative	1	2				
Sang	2	Vegetative	1	2				
		Generative	0	1				
Total		2	42					
Totally		11	109					

Table 2. Recapitulation	of Hypersensitive	Test of isolates	of PSB from	n isolation in	upland rice	e cultivation	areas in
Bangka Regency.							

Location	Pastoria	Hypersensitive test			
Location	Dacteria	Vegetative phase	Generative phase		
Davahanya	Rhizosphere	9	19		
rayabenua	Endophytes	4	4		
	Total	13	23		
Saina	Rhizosphere	27	0		
Saling	Endophytes	1	0		
	Total	28	0		
	Totaly	4.1	$2\overline{3}$		

Ability of isolates in dissolving phosphate

Testing the capacity to dissolve P was conducted on 23 bacterial isolates. The ability to dissolve P was examined in a total of 23 PSB isolates, as presented in Table 3. The dissolved P values varied among the 23 isolates, ranging from 9 to 200 mg L-1 in AlPO4 liquid media and 3.8 to 843 mg L⁻¹ in $Ca_3(PO_4)_2$ liquid media. The solubility value of P in AlPO₄ and Ca₃(PO₄)₂ liquid media exhibited a range of 0.0013 to 0.0344% and 0.0008 to 0.1842%, respectively.

Testing of Organic Acids Content, Enzyme Phosphatase and Molecular Identification of Selected Bacteria Isolates

Phosphatase enzymes and organic acids were measured in the 6 selected bacterial isolates that exhibited significantly dissolved P value and particularly high solubility in AlPO4 medium. These selected isolates were identified as BEP1V4, BEP1V7, BEP2G15, BEP2G18, BEP2V11, and BRP2V6 (Table 4), originating from Payabenua Village. BEP1V4 and BEP1V7 came from location 1, namely the upland rice cultivation area on new openings, while BEP2G15, BEP2G18, BEP2V11, and BRP2V6 were derived from the rice field cultivation area superimposed with pepper plants. BEP2G15 and BEP2G18 isolates were in the generative growth phase, while the others were in the vegetative phase.

Molecular identification results showed that BEP1V4, BEP1V7, BEP2G15, and BEP2G18 were the genus Burkholderia, while BEP2V11 and BRP2V6 were the genus Serratia as shown in Table 5 and Figure 3.

Discussion

The dominant bacteria obtained from isolation are endophytic PSB isolates derived from the root of the upland rice plant. Hartanti (2020) also succeeded in isolating four types of endophytic bacteria from the roots of the Situbagendit variety rice plant, while Putriani et al. (2019) isolated

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N		PSI _{Ca} -	Disso	olved P	Solubility P		
NO	Isolate Code		$AlPO_4$	$(Ca_3PO_4)_2$	$AlPO_4$	$(Ca_3PO_4)_2$	
			mg L-1		%		
1	BEP_1V_3	0.24	12.5	5.8	0.0021	0.0013	
2	BEP_1V_4	0.25	17.3	48	0.0030	0.0105	
3	BEP_1V_7	0.27	17.8	54	0.0031	0.0118	
4	BEP_1V_9	0.27	7.8	4.3	0.0013	0.0009	
5	BEP_1V_{10}	0.40	13	6.3	0.0022	0.0014	
6	BEP_1V_{12}	0.34	11.5	811	0.0020	0.1772	
7	BEP_1V_{13}	0.45	14.8	18.3	0.0025	0.0040	
8	BEP_1V_{14}	0.68	14.8	19.8	0.0025	0.0043	
9	BEP_1V_{16}	0.24	14	39	0.0024	0.0085	
10	BEP_2G_1	0.26	15.5	9	0.0027	0.0020	
11	BEP_2G_2	0.15	16.3	9.3	0.0028	0.0020	
12	BEP_2G_3	0.27	9	8.5	0.0015	0.0019	
13	BEP_2G_4	0.18	13.3	5.8	0.0023	0.0013	
14	BEP_2G_5	0.25	17.8	9.3	0.0031	0.0020	
15	BEP_2G_6	0.42	17.8	11.5	0.0031	0.0025	
16	BEP_2G_{15}	0.14	200	843	0.0344	0.1842	
17	BEP_2G_{18}	0.22	26.3	11	0.0045	0.0024	
18	BEP_2V_1	0.52	16.3	13.3	0.0028	0.0029	
19	$\mathrm{BEP}_{2}\mathrm{V}_{4}$	0.30	16	6.3	0.0027	0.0014	
20	BEP_2V_9	0.32	14.5	14.5	0.0025	0.0032	
21	BEP_2V_{11}	0.15	47	378	0.0081	0.0826	
22	BEP_2V_{15}	0.45	26.3	3.8	0.0045	0.0008	
23	BRP_2V_6	-	22	367	0.0038	0.0802	

Table 3. The ability of selected isolates to dissolve phosphates from upland rice cultivation in Bangka Regence

Table 4. The content of phosphatase enzymes and organic acids of PSB from upland rice cultivation in Bangka Regency.

		Phosphatase			Organic acids		
No	Isolate code	Enzyme (ppm)	Acetic acid	Lactic acid	Malic acid	Citric acid	Oxalic acid
		ppm		mg L-	1		
1	BEP_1V_4	1.36	3.98	nm	nm	0.251	0.373
2	BEP_1V_7	2.63	2.026	1.058	nm	0.063	0.139
3	BEP_2G_{15}	2.43	3.292	nm	nm	0.173	0.343
4	BEP_2G_{18}	2.69	4.35	0.655	1.456	0.211	0.335
5	BEP_2V_{11}	2.04	2.372	1.00	0.455	0.078	0.202
6	BRP_2V_6	0.90	0.493	0.907	0.921	0.088	nm

ppm=part per million, nm= not measured

six endophytic isolates from rice roots in the Aceh area. According to Ji et al. (2014), endophytic bacteria in rice plants had a fairly important role in spurring plant growth. Marwan et al. (2021) reported that the isolates from local Jambi rice varieties had the potential to be developed as biological control agents against blast disease caused by *P. oryzae.* Meanwhile, Prihatiningsih et al. (2021) concluded that three endophytic bacteria (SM1, SB 1, and SB 3) associated with rice roots could be categorized as potential plant growth promoters.

The population of endophytic bacteria was found in Payabenua when compared to the Saing area. In the Payabenua area, bacteria were isolated from rice fields cultivated in dry areas. In Saing area, the rice field area is pure upland area since the land become flooded when rainy season. Therefore, the conditions of the different planting areas caused the variety of isolates found. Bacterial isolates from the Payabenua area

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Tabl	e 5. Molecular id	entification of selected isolates based o	on 16 Sr DNA sequence similarity.
No	Isolate code	Bacterial name	Similarity index
1	BEP_1V_4	Bukholderia sp	Homology 99,92% with <i>Burkholderia</i> sp. strain RB141 16S ribosomal RNA gene, partial se-
			quence.
2	BEP_1V_7	Bukholderia cenocepcia	Homology 99,93% with <i>Burkholderia cenocepa-</i> <i>cia</i> strain PC184 Mulks chromosome 3, com-
			plete sequence.
3	BEP_2G_{15}	Bukholderia latens	Homology 99,78% with <i>Burkholderia latens</i> strain AU17928 chromosome 1, complete sequence.
4	BEP_2G_{18}	Burkholderia vietnamiensis	Homology 99,57% with <i>Burkholderia viet-</i> namiensis strain TVV75, partial sequence
5	BEP_2V_{11}	Serratia marcescens	Homology 99,86%, with <i>Serratia marcescens</i> strain CTC639-K12, partial sequence.
6	BRP_2V_6	Serratia surfactantfaciens	Homology 99,81% with Serratia surfactantfa- ciens strain YD25.



Figure 3. The six selected isolated from Upland Rice Cultivation Areas in Bangka Regency.

were predominantly reported in the vegetative growth phase and came from rice field cultivation areas superimposed with pepper plants. Pande et al. (2020) stated that physical and chemical characteristics of the soil, organic matter, phosphorus concentration, and cultural activities had an impact on the PSB population.

Despite the HDB prevalence in the vegetative growth phase, dominant selected isolates originated from the generative growth phase. However, the measurement of the phosphatase enzyme as well as the levels of organic acids obtained from the generative phase were relatively higher than the vegetative phase. According to Setiawati et al. (2014), the ability to produce enzyme phosphatase and the production of organic acids is a characteristic of the ability of PSB isolates. According to Ranjan et al. (2013), a phosphatase is a group of enzymes that catalyzes an enzymatically hydrolytic mineralization reaction with the release of undissolved phosphates into dissolved. Situmorang et al. (2015) explained that the higher the enzyme activity produced by PSB, the greater clear zone. Fitriatin et al. (2020) stated that the important role of the phosphatase enzyme was in the hydrolysis process of organic phosphate into inorganic phosphate.

In addition to the production of phosphatase enzymes, PSB also generated organic acids, which played a crucial role in the P solubilization. These organic acids contributed to the process through several mechanisms by acidifying the rhizosphere, aiding in the chelation of cations responsible for phosphate precipitation, facilitating the availability of metal ions associated with insoluble Ca, Al, and Fe phosphate compounds, and competing with P for adsorption sites on the soil. (Kishore et al. 2015). Organic acids in PSB include citric, gultamic, succinic, oxalic, malic, fumaric, and tartaric acids (Seshachala & Tallapragada 2012; Asrul & Aryantha 2020). Malic, citric and oxalic acids have low molecular weight and are effective in alkalizing Al (Hafif et al. 2010). Bacterial isolates of the *Burkholderia* sp group also produced low molecular weight organic acids compared to *Serratia* sp. Therefore, the solubilization ability of *Burkholderia* sp. group is more effective in Al medium compared to the *Serratia* sp. Fitriatin et al. (2021) reported that organic acids (lactate, citrate, oxalate, and tartrate) in *Burkholderia* sp. bacteria (WK strain 11 and MQ-14W strain) were produced more at pH 4.5 than at pH 7 or 10.5.

Genus of endophytic bacteria that dominate the internal tissues of rice plants are *Pseudomonas*, *Bacillus*, *Streptomyces*, *Azospirillum*, and *Azoto*bacter (Kumar et al. 2020). Furthermore, Burkholderia is a common bacterial family involved in the solubilization of phosphate (Kishore et al. 2015). Raweekul et al. (2016) isolated endophytic bacteria from rice and reported that the genus Burkholderia was dominant on the stalks. However, Aroumougame's (2020) study found that Burkholderia was dominant in the roots of rice plants. Several studies related to the genus in food crops have been reported, such as Burkholderia caribensis of rice agroecosystems of South Assam, India (Roy et al. 2013) and Burkholderia cepacia in maize (Zhao et al. 2014). The genus consists of more than 40 bacteria that cluster to form a species complex known as *B. cepacia* (Bcc) (Arriel-Elias et al. 2019) Burkholderia is a bifunctional genus because some of its species establish symbiotic-mutualistic relationships with plants, and symbiotic-pathogenic associations with plants, animals, and humans (Espinosa-Victoria et al. 2020).

The phosphate-solubilizing bacteria obtained in this study are local bacterial isolates, so they have prospects for development in the Bangka region in particular. In addition, generally, the bacteria obtained are endophytic bacteria, so it is possible to use them on other crops besides rice. Allegedly, besides having a mechanism as a phosphate solubilizer, the bacterial isolates obtained have other functions as biocontrol as well as the ability to PGPR. Therefore, there is a need for further research to find out some information related to the bacteria obtained in this study.

CONCLUSIONS

In conclusion, a total of 6 isolates of potential PSB were obtained from the ultisol land of upland rice cultivation in Payabenua Village, Bangka Regency. Among these isolates, *Bukholderia* sp., *Bukholderia cenocepcia*, *Bukholderia latens*, and *Burkholderia vietnamiensis* belonged to the genus *Burkholderia*. Furthermore, the remaining two were identified as *Serratia marcescens*, belonging to the genus *Serratia*. The levels of both phosphatase enzymes and organic acids produced by the genus *Burkholderia* were found to be higher than those produced by *Serratia*. The solubility value of P in AlPO₄ and Ca₃(PO₄)₂ liquid media exhibited a range of 0.0013 to 0.0344% and 0.0008 to 0.1842%, respectively.

AUTHOR CONTRIBUTION

KK designed the study process, collected and analyzed the data, as well as wrote the manuscript. MRS designed, supervised the study, and wrote the manuscript. AB, ERP, and SI supervised the study and edited the manuscript.

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CONFLICT OF INTEREST

The authors declare no conflict of Interest

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