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RESEARCH ARTICLE

Phosphorus-solubilizing bacterial endophytes, *Burkholderia silvatlantica* L-VT09 and N-VT06, in improving characteristics of acid sulfate soil and pineapple yield

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ABSTRACT

Acid sulfate soils are limiting the productivity of pineapples (*Ananas comosus* (L.) Merr. var. *comosus*) by lacking available P content. This study aims to evaluate the potency of P-solubilizing bacterial endophytes, *Burkholderia silvatlantica* L-VT09 and N-VT06, on acid sulfate soil properties and features of pineapples grown in that soil. The two-factor experiment was arranged according to a completely randomized block design. One factor was P fertilizer levels: 25% P, 50% P, 75% P, and 100% P; the other was supplementations of P-solubilizing bacterial endophytes: No supplementations, individual supplementation of L-VT09, individual supplementation of N-VT06, and mixed supplementation of both L-VT09 and N-VT06. The result revealed that supplying the mixture of *B. silvatlantica* L-VT09 and N-VT06 did improve the soil fertility as increasing available P content and bacterial density (12.6 mg P kg^{-1} and 3.82 most probable number g^{-1} dry soil weight, respectively), P uptake (19.2 kg ha^{-1}), growth as boosting plant height (8.30 cm), leaves number per plant (6.60 leaves $plant^{-1}$), D-leaf length and width (2.80 and 0.32 cm, correspondingly), stem size (3.40 cm in length and 0.44 cm in diameter), yield components as enhancing fruit length and fruit diameter (3.50 and 1.46 cm, sequentially), yield (11.5 t ha^{-1}), and pineapple fruit quality as ameliorating sugar content (1.71 °Brix) and water content (166.4 mL), in comparison with the no bacteria supplied case. Supplying the mixture of *B. silvatlantica* L-VT09 and N-VT06 altered 75% P but still maintained pineapple yield.

Keywords: Acid sulfate soil, *Ananas comosus* var. *comosus*, bacterial endophytes, *Burkholderia silvatlantica*, pineapple, phosphorous-solubilizing.

INTRODUCTION

Phosphorous (P) is one of the most essential macronutrients, playing an important role in plant development (Divjot et al., 2021). However, the cycle of P is different from other important nutrients, such as N and C, because it does not go through gaseous forms, such as N_2 or CO_2 (Percival et al., 2020). Furthermore, P within agricultural soils is usually limited due to bonds between P and cations, creating insoluble compounds (Elhaissoufi et al., 2021). Strengite ($FePO_4 \cdot 2H_2O$) and variscite ($AlPO_4 \cdot 2H_2O$), tricalcium phosphate ($Ca_3(PO_4)_2$), dicalcium phosphate ($CaHPO_4$), dicalcium phosphate dihydrate ($CaHPO_4 \cdot H_2O$), fluorapatite ($Ca_5(PO_4)_3F$), hydroxyapatite ($Ca_5(PO_4)_3OH$), and octacalcium phosphate ($Ca_4H(PO_4)_3 \cdot 2\text{-}5H_2O$) have been found to be common P minerals (Yadav and Verma, 2012). Although fertilizing chemical P is advised to manage soil P deficiency, the usage potency of chemical P in soils is low, only 10%-25% (Etesami, 2020), which restricts providing available P for plants (Etesami and Jeong, 2021). Nevertheless, using chemical P fertilizers over recommendations affects the environment (Etesami and Jeong, 2021) and may be immobilized by oxides of Al, Fe, and Ca in soils that plants are unable to take (Divjot et al., 2021). Thus, many approaches have been applied to solubilize soil-insoluble P for plants to absorb. Among these approaches, utilizing P-solubilizing bacterial endophytes is a promising one (Elhaissoufi et al., 2021). Some P-solubilizing bacteria, such as *Arthrobacter*, *Bacillus*, *Burkholderia*, *Pseudomonas*, *Rhizobium*, and *Serratia*, have been determined to be beneficial to plants via enhancing P use efficiency (Divjot et al., 2021). According to Kim et al. (2021), P-solubilizing bacteria, the *Methylobacterium* sp. strain PS and *Caballeronia* sp. strain EK ameliorated the growth of tomatoes in acid sulfate soil. A strain of *B. silvatlantica* has been proven as an endophyte of pineapples (da Silva et al., 2020). Bacterial endophytes are known to be capable of enhancing the growth and development of plants (Santoyo et al., 2016). The P-solubilizing bacteria have the potential to be used as a biofertilizer, stimulating the development of plants grown in the acid sulfate soil by improving the P uptake (Kim et al., 2021). Biofertilizers are an environmentally friendly and sustainable approach to alter chemical P fertilizer (Chandran et al., 2021) and are an option for maintaining the available P content in soils (Flatian et al., 2021). Some bacterial strains have been applied as biofertilizers and are commercialized industrially, such as *Bacillus*, *Pseudomonas*, *Enterobacter*, and *Azospirillum* (Rai et al., 2020). In the study by Rana et al. (2021), some endophytic bacteria, such as *Pseudomonas brenneri*, *Ewingella americana*, and *Pantoea agglomerans*, have been found to be capable of solubilizing P. Etesami and Jeong (2021) reported that P-solubilizing bacteria improve soil fertility, enhancing P availability and P uptake in plants. Thus, the application of P-solubilizing bacteria can help reduce the chemical P fertilizer while still supporting crop growth and yield (Rafi et al., 2019) and also soil fertility (Dhankhar et al., 2013). Therefore, this study was conducted to determine the efficiency of P-solubilizing bacterial endophytes, the *Burkholderia silvatlantica* L-VT09 and N-VT06, on improvements of an acid sulfate soil fertility, and the P uptake, growth, yield, and fruit quality of pineapples cultivated in the soil as well. The bacteria mixture was hypothesized to reduce the applied chemical P fertilizer and positively affect the fertility of acid sulfate soil and the growth and yield of pineapples.

MATERIALS AND METHODS

The experiment was conducted in an acid sulfate soil, whose properties are shown in Table 1 from February 2021 to June 2022. The "Queen" pineapple (*Ananas comosus* (L.) Merr. var. *comosus*) was selected for the experiment, with supplements from biofertilizers and chemical fertilizers. Strains of P-solubilizing bacterial endophytes *Burkholderia silvatlantica* L-VT09 and N-VT06 were isolated and screened from roots and stems of pineapple plants cultivated in the acid sulfate soil (Khuong et al., 2022) and stored at -80 °C in the Faculty of Crop Science, College of Agriculture, Can Tho University. The two strains had been activated before being used. Types of chemical fertilizers used for pineapple included urea (46.3% N), superphosphate (16% P_2O_5), and potassium chloride (60% K_2O), with commercial names of Phu My, Long Thanh, and Phu My, respectively.

Experimental design

The experiment had two factors and followed a randomized complete block design. In detail, the first factor (A) was P fertilizer levels and consisted of four levels of P: 25%, 50%, 75%, and 100% to create a series of changing amounts of P fertilizer used according to the recommended fertilizer formula; the second factor (B) was different compositions of supplying the P-solubilizing bacteria, including the control: no bacteria used, single strain of *B. silvatlantica* L-VT09, single strain of *B. silvatlantica* N-VT06, and a mixture of L-VT09 and N-VT06. Each treatment contained four replicates corresponding to four experimental plots, each of which was 25 m^2 width.

The fertilizer formula for pineapple was 10 N-7 P_2O_5-8 K_2O (g plant^{-1}), according to the study by Be and Hoa (2009).

The bacteria were supplied into the ground of pineapple at a density of 2.5×10^8 CFU mL^{-1}. The ground of pineapple was evenly irrigated with bacteria suspensions five times at 60, 105, 150, 195, and 240 d after planting at 6.25×10^5 CFU plant^{-1} (after chemical fertilizers had been applied).

Table 1. Characteristics of acid sulfate soil for pineapple. EC: Electrical conductivity.

Characteristics	Value
EC, mS cm^{-1}	0.67
pH_{H2O}	2.96
pH_{KCl}	2.79
NH_4^+, mg NH_4^+ kg^{-1}	79.50
NO_3^-, mg NO_3^- kg^{-1}	56.50
$P_{available}$,	144.60
Fe-P, mg P kg^{-1}	488.60
Al-P, mg P kg^{-1}	97.70
Ca-P, mg P kg^{-1}	35.10
N_{total}, % N	0.23
P_{total}, % P	0.15

Criteria of evaluation

According to Sparks et al. (1996), soil analysis was proposed on pH_{H2O}, pH_{KCl}, electrical conductivity (EC), total P content, available P content, ferric phosphate (Fe-P), aluminum phosphate (Al-P), calcium phosphate (Ca-P), NH_4^+ concentration, NO_3^- concentration, and total N content. Bacterial densities in the soil were measured following the Most Probable Number (MPN) method in the NBRIP medium, as described by Naoki et al. (2001).

The P concentrations in the crown, flesh, core, shell, slip, peduncle, stem, and leaves were analyzed according to Houba et al. (1988).

Growth features were determined at harvesting when 20 plants were randomly collected from each treatment and measured following the method of Huu et al. (2022).

Twenty fruits were randomly derived from each treatment and recorded for yield components, yield, and fruit quality analysis at harvesting. Fruit diameter (cm), fruit length (cm), fruit weight (g), and yield (t ha^{-1}) were calculated according to Huu et al. (2022). Fruit quality parameters, namely, water content (mL), juice pH, and fruit color, were measured following the method of Huu et al. (2022). The total acid content (mg L^{-1}) measurement followed the method of Guerrant et al. (1982); sugar content (°Brix) was measured according to Saini et al. (2001).

Statistical analysis

A two-way ANOVA was run using the general linear model procedure to calculate the effects of P fertilizer levels (A), the supplementations of bacteria (B), and the interaction between them. The means of separation of P fertilizer levels and the supplementations of bacteria for the different parameters were performed by Duncan's post hoc test at 5% significance. Data were analyzed using the SPSS 13.0 statistical package (IBM, Armonk, New York, USA).

RESULTS

Effects on the fertility of acid sulfate soil for pineapples

Results in Table 2 show different nonsignificant values of pH_{H2O}, pH_{KCl}, EC, total P content, and total N content between levels of P fertilizer and bacteria supplementations, which were correspondingly 2.51-2.78, 2.33-2.56, 1.75-2.15 mS cm^{-1}, 0.086%-0.109% P and 0.215%-0.239% N.

Table 2. Effects of P-solubilizing bacterial endophytes, *Burkholderia silvatlantica* L-VT09 and N-VT06, on the fertility of acid sulfate soil for pineapple. Numbers followed by different letters are significantly different. *Different at 5% significance; ns: nonsignificant. EC: electrical conductivity; MPN: most probable number; DSW: dry soil weight; A: fertilizer factor; B: bacteria factor; L-VT09: supplying only the P-solubilizing *B. silvatlantica* L-VT09 strain; N-VT06: supplying only the P-solubilizing *B. silvatlantica* N-VT06; L-VT09 + N-VT06: supplying the mixture of P-solubilizing *B. silvatlantica* L-VT09 and N-VT06.

Factors		pH_{H2O}	pH_{KCl}	EC	P_{total}	$P_{available}$	Fe-P	Al-P	Ca-P	N_{total}	NH_4^+	NO_3^-	Log bacterial density
				mS cm^{-1}	% P	mg P kg^{-1}				% N	mg NH_4^+ kg^{-1}	mg NO_3^- kg^{-1}	MPN g^{-1} DSW
Level of P fertilizer (A)	100% P	2.59	2.37	2.15	0.093	44.6^{a}	231.7^{a}	65.1^{a}	37.0^{a}	0.216	52.0^{a}	6.13	5.34
	75% P	2.64	2.45	2.00	0.101	42.1^{b}	204.6^{b}	52.7^{b}	33.0^{b}	0.215	50.3^{a}	6.06	5.26
	50% P	2.55	2.40	1.96	0.098	39.8^{c}	192.4^{c}	37.3^{c}	29.9^{c}	0.239	42.1^{b}	5.80	5.31
	25% P	2.78	2.56	1.75	0.095	23.5^{d}	147.8^{d}	28.3^{d}	26.2^{d}	0.218	35.5^{c}	5.58	5.27
Supplementation of bacteria (B)	No bacteria	2.78	2.53	2.03	0.095	29.2^{c}	253.2^{a}	52.6^{a}	35.7^{a}	0.231	37.7^{d}	5.15^{b}	2.62^{c}
	L-VT09	2.72	2.54	1.81	0.109	39.8^{b}	208.3^{b}	42.8^{c}	31.7^{b}	0.219	53.2^{a}	6.37^{a}	6.09^{b}
	N-VT06	2.51	2.33	2.00	0.096	39.3^{b}	156.2^{c}	42.5^{c}	29.7^{c}	0.222	43.0^{c}	6.08^{a}	6.03^{b}
	L-VT09 + N-VT06	2.56	2.38	2.02	0.086	41.8^{a}	158.8^{c}	45.3^{b}	29.0^{c}	0.215	46.0^{b}	5.98^{a}	6.44^{a}
P (A)		ns	ns	ns	ns	*	*	*	*	ns	*	ns	ns
P (B)		ns	ns	ns	ns	*	*	*	*	ns	*	*	*
P (A×B)		ns	ns	ns	ns	*	*	*	ns	ns	*	ns	ns
CV, %		12.2	12.2	23.0	46.2	6.5	3.5	6.5	8.2	18.2	7.45	11.1	1.75

Table 2 illustrates that the treatments fertilized with 100% and 75% P had equivalent amounts of NH_4^+, with 52.0 and 50.3 mg NH_4^+ kg^{-1}, higher than those in the treatment fertilized with 25% P, with 35.5 mg NH_4^+ kg^{-1}. The treatment supplied with the single strain of *B. silvatlantica* L-VT09 possessed the highest concentration of NH_4^+ (53.2 mg NH_4^+ kg^{-1}), and the lowest result appeared in the treatment with bacteria supplied (37.7 mg NH_4^+ kg^{-1}). Contents of NO_3^- varied no significantly between levels of P fertilizer applied and ranged from 5.58 to 6.13 mg NO_3^- kg^{-1} and were different ($P < 0.05$) for the bacteria factor, with 5.98-6.37 mg NO_3^- kg^{-1} in the treatments supplied with bacteria and 5.15 mg NO_3^- kg^{-1} in the treatment without ones.

Bacterial densities between levels of P fertilizer were no significantly different and fluctuated roughly 5.26-5.34 MPN g^{-1} dry soil weight (DSW). The treatment supplied with the mixture of the *B. silvatlantica* L-VT09 and N-VT06 strains showed the highest bacterial density, with 6.44 MPN g^{-1} DSW. Bacterial densities in the treatment supplied with the single strain of L-VT09 and in the one supplied with the single strain of N-VT06 were statistically the same and higher than that in the treatment without bacteria supplied, i.e., 6.03-6.09 compared to 2.62 MPN g^{-1} DSW (Table 2).

Effects on P uptake of pineapples in acid sulfate soil

Dry biomass in stovers of pineapples in acid sulfate soil. Table 3 indicates that dry biomass in the crown, flesh, core, shell, slip, peduncle, stem, and leaves changed under the influences of P fertilizer levels and bacteria supplementations (P < 0.05). In particular, decreasing the P fertilizer level led to reductions in the dry biomass in crown, flesh, core, shell, and slip, with results at 468.1 > 456.8 > 445.3 > 359.7 kg ha^{-1}, 1692.7 > 1419.8 > 1193.8 > 1031.6 kg ha^{-1}, 432.8 > 402.9 > 351.7 > 265.2 kg ha^{-1}, 1790.5 > 1559.3 > 1389.5 > 1243.6 kg ha^{-1}, and 844.1 > 774.1 > 667.3 > 606.3 kg ha^{-1}, respectively, corresponding to P fertilizer levels at 100% > 75% > 50% > 25% P. In addition, the dry biomass in peduncle and stem were equivalent between the treatments fertilized with 100% and 75% P and higher than those in the treatments fertilized with 50% and 25% P, with 394.8-405.8 compared with 338.7-363.5 kg ha^{-1} and 1095.4-1105.6 compared with 948.9-1050.6 kg ha^{-1}, respectively. In the treatment fertilized with 100% P, the dry biomass in leaves was higher than that in the treatments fertilized with 50% and 25% P (5178.0 > 4347.5 ~ 4261.3 kg ha^{-1}). Dry biomass in stovers of pineapple in the treatment without bacteria was lower than that in the treatments supplied with bacteria, 321.6 compared to 413.8-546.3 kg ha^{-1} in the crown, 954.9 compared with 1269.8-1703.6 kg ha^{-1} in the flesh, 264.9 compared with 382.5-405.7 kg ha^{-1} in the core, 1150.8 compared with 1460.6-1829.0 kg ha^{-1} in the shell, 502.2 compared with 624.2-1001.7 kg ha^{-1} in slips, 318.3 compared with 365.9-431.7 kg ha^{-1} in the peduncle, 889.9 compared with 1005.4-1157.9 kg ha^{-1} in the stem, and 3923.1 compared with 4733.4-5335.1 kg ha^{-1} in the slip.

Table 3. Effects of P-solubilizing bacterial endophytes, *Burkholderia silvatlantica* L-VT09 and N-VT06, on dry biomass in stovers of pineapple in acid sulfate soil. Numbers followed by different letters are significantly different. *Different at 5% significance; A: fertilizer factor; B: bacteria factor; L-VT09: supplying only the P-solubilizing *B. silvatlantica* L-VT09 strain; N-VT06: supplying only the P-solubilizing *B. silvatlantica* N-VT06; L-VT09 + N-VT06: supplying the mixture of P-solubilizing *B. silvatlantica* L-VT09 and N-VT06.

Factors		Crown	Flesh	Core	Shell	Slips	Peduncle	Stem	Leaves
		kg ha^{-1}							
Level of P fertilizer (A)	100% P	468.1^a	1692.7^a	432.8^a	1790.5^a	844.1^a	405.8^a	1095.4^a	5178.0^a
	75% P	456.8^b	1419.8^b	402.9^b	1559.3^b	774.1^b	394.8^a	1105.6^a	4944.8^b
	50% P	445.3^c	1193.8^c	351.7^c	1389.5^c	667.3^c	363.5^b	1050.6^b	4347.5^c
	25% P	359.7^d	1031.6^d	265.2^d	1243.6^d	606.3^d	338.7^c	948.9^c	4261.3^c
Supplementation of bacteria (B)	No bacteria	321.6^d	954.9^d	264.9^c	1150.8^d	502.2^d	318.3^d	889.9^c	3923.1^c
	L-VT09	546.3^a	1269.8^c	399.5^a	1460.6^c	624.2^c	365.9^c	1005.4^b	4739.9^b
	N-VT06	413.8^c	1409.7^b	382.5^b	1542.4^b	763.6^b	386.9^b	1147.3^a	4733.4^b
	L-VT09 + N-VT06	448.2^b	1703.6^a	405.7^a	1829.0^a	1001.7^a	431.7^a	1157.9^a	5335.1^a
P (A)		*	*	*	*	*	*	*	*
P (B)		*	*	*	*	*	*	*	*
P (A×B)		*	*	*	*	*	*	*	*
CV, %		3.52	6.52	4.94	4.04	3.76	4.23	5.22	3.51

Phosphorus concentrations in stovers of pineapples in acid sulfate soil

The results in Table 4 show that P contents in pineapple stovers were different between levels of P fertilizer and supplementations of bacteria (P < 0.05). To be more specific, the treatment fertilized with 100% P reached higher concentrations of P in the crown and leaves than the treatments fertilized with 50% and 25% P, ranking in order of 0.576% > 0.509% ~ 0.492% and 0.367% > 0.326% ~ 0.310%, respectively. P contents in flesh and slips were at the highest point in the treatment fertilized with 100% P (0.330% and 0.560%, respectively), at the lowest point in the treatment fertilized with 25% P (0.271% and 0.458%, respectively) and those in the treatments fertilized with 75% and 50% P were statistically the same (0.308% ~ 0.292% and 0.516% ~ 0.503%). The P concentrations in core and shell progressively based on the levels of P fertilizer applied from 100% > 75% > 50% > 25% P and valued at 0.245% > 0.228% > 0.209% > 0.182% and 0.257% > 0.237% > 0.224% > 0.201%. The treatments fertilized with 100% and 75% P possessed higher P content in the peduncle and stem than those in the treatments fertilized with 50% and 25% P; i.e., they were ranked, according to a decrease in P fertilizer levels, 0.222% ~ 0.214% > 0.184%-0.194% and 0.506% ~ 0.514% > 0.471%-0.445%. The treatments supplied with the mixture of *B. silvatlantica* L-VT09 and N-VT06 strains, with only the L-VT09 strain and with only the N-VT06 strain, all had P concentrations that were higher than those in the treatment without bacteria supplied in the crown (0.508%-0.572% compared with 0.482%), in the flesh (0.299%-0.326% compared with 0.276%), in core (0.220%-0.228% compared with 0.192%), in the shell (0.230%-0.251% compared with 0.206%), in slips (0.521%-0.532% compared with 0.454%), in stem (0.458%-0.551% compared with 0.424%), and in leaves (0.340%-0.350% compared with 0.310%). However, in the treatments supplied the mixture of the L-VT09 and N-VT06 strains and with only the N-VT06 strain, the P contents in peduncle (0.206%-0.235%) were higher than in the treatments supplied with only the L-VT09 strain and in the one without bacteria (0.188%-0.186%).

Table 4. Effects of P-solubilizing bacterial endophytes, *Burkholderia silvatlantica* L-VT09 and N-VT06, on P concentrations in stovers of pineapple in acid sulfate soil. Numbers followed by different letters are significantly different. [*]Different at 5% significance; [ns]: nonsignificant; A: fertilizer factor; B: bacteria factor; L-VT09: supplying only the P-solubilizing *B. silvatlantica* L-VT09 strain; N-VT06: supplying only the P-solubilizing *B. silvatlantica* N-VT06; L-VT09 + N-VT06: supplying the mixture of P-solubilizing *B. silvatlantica* L-VT09 and N-VT06.

Factors		Crown	Flesh	Core	Shell	Slips	Peduncle	Stem	Leaves
		%							
Level of P fertilizer (A)	100% P	0.576^a	0.330^a	0.245^a	0.257^a	0.560^a	0.222^a	0.514^a	0.367^a
	75% P	0.541^b	0.308^b	0.228^b	0.237^b	0.516^b	0.214^a	0.506^a	0.346^b
	50% P	0.509^c	0.292^b	0.209^c	0.224^c	0.503^b	0.194^b	0.471^b	0.326^c
	25% P	0.492^c	0.271^c	0.182^d	0.201^d	0.458^c	0.184^c	0.445^b	0.310^c
Supplementation of bacteria (B)	No bacteria	0.482^c	0.276^c	0.192^b	0.206^c	0.454^b	0.186^c	0.424^d	0.310^b
	L-VT09	0.508^b	0.300^b	0.220^a	0.230^b	0.530^a	0.188^c	0.458^c	0.349^a
	N-VT06	0.557^a	0.299^b	0.225^a	0.232^b	0.521^a	0.206^b	0.502^b	0.340^a
	L-VT09 + N-VT06	0.572^a	0.326^a	0.228^a	0.251^a	0.532^a	0.235^a	0.551^a	0.350^a
P (A)		*	*	*	*	*	*	*	*
P (B)		*	*	*	*	*	*	*	*
P (A×B)		ns	ns	ns	*	*	ns	ns	ns
CV, %		6.68	7.54	7.38	6.69	5.28	5.97	8.99	6.69

Phosphorus uptakes in stovers of pineapples in acid sulfate soil

Table 5 shows that P uptakes in stovers of plants were different between P fertilizer levels and bacteria supplementations ($P < 0.05$). In detail, P uptakes in crown, flesh, core, shell, slips, peduncle, and leaves went down, corresponding to P fertilizer levels drop from 100%, 75%, 50% to 25% P, from 2.70 > 2.48 > 2.28 > 1.78 kg ha^{-1} in crown, 5.63 > 4.46 > 3.52 > 2.83 kg ha^{-1} in flesh, 1.070 > 0.930 > 0.740 > 0.494 kg ha^{-1} in core, 4.65 > 3.72 > 3.14 > 2.52 kg ha^{-1} in shell, 4.76 > 4.06 > 3.42 > 2.77 kg ha^{-1} in slips, 0.915 > 0.857 > 0.711 > 0.623 kg ha^{-1} in peduncle, and 19.1 > 17.1 > 14.2 > 13.3 kg ha^{-1} in leaves. Besides, P uptakes in the stem were significantly unchanged between the treatments fertilized with 100% and 75% P. However, they were higher than in the treatments fertilized with 50% and 25% P, 5.69 ~ 5.67 compared with 4.24-4.99 kg ha^{-1}, respectively. The treatments supplied with bacteria had higher P uptakes in stovers than those in the treatment without bacteria. In particular, for P uptakes in crown, flesh, core, shell, slips, peduncle, stem, and leaves, the results were 2.32-2.78 > 1.56, 3.89-5.61 > 2.68, 0.870-0.937 > 0.526, 3.41-4.64 > 2.38, 2.34-5.34 > 2.29, 0.692-1.024 > 0.594, 4.60-6.44 and 16.2-18.8 > 12.2 kg ha^{-1}, respectively.

In Table 5, the treatment fertilized with 100% P had the highest total P uptake (44.5 kg ha^{-1}) and the treatment fertilized with 25% P had the lowest one (28.6 kg ha^{-1}). The treatment supplied with the mixture of *B. silvatlantica* L-VT09 and N-VT06 strains had the highest result, the second highest was in the treatment supplied with only the N-VT06 strain, and the treatments supplied with only L-VT09 strain and without bacteria appeared as the lowest one (45.3 > 37.8 > 36.1 > 26.1 kg ha^{-1}). The two factors, bacteria supplementations and levels of P fertilizer, interacted with each other in P uptakes in flesh, core, shell, slips, peduncle, stem, and leaves and total P uptake ($P < 0.05$). Nevertheless, the interaction in the P uptake in the crown was insignificant.

Table 5. Effects of P-solubilizing bacterial endophytes, *Burkholderia silvatlantica* L-VT09 and N-VT06, on P uptake in stovers of pineapple in acid sulfate soil. Numbers followed by different letters are significantly different. *Different at 5% significance; ns: nonsignificant; A: fertilizer factor; B: bacteria factor; L-VT09: supplying only the P-solubilizing *B. silvatlantica* L-VT09 strain; N-VT06: supplying only the P-solubilizing *B. silvatlantica* N-VT06; L-VT09 + N-VT06: supplying the mixture of P-solubilizing *B. silvatlantica* L-VT09 and N-VT06.

Factors		Crown	Flesh	Core	Shell	Slips	Peduncle	Stem	Leaves	Total P
		kg ha^{-1}								
Level of P fertilizer (A)	100% P	2.70^a	5.63^a	1.070^a	4.65^a	4.76^a	0.915^a	5.69^a	19.1^a	44.5^a
	75% P	2.48^b	4.46^b	0.930^b	3.72^b	4.06^b	0.857^b	5.67^a	17.1^b	39.3^b
	50% P	2.28^c	3.52^c	0.740^c	3.14^c	3.42^c	0.711^c	4.99^b	14.2^c	33.0^c
	25% P	1.78^d	2.83^d	0.494^d	2.52^d	2.77^d	0.623^d	4.24^c	13.3^d	28.6^d
Supplementation of bacteria (B)	No bacteria	1.56^d	2.68^d	0.526^c	2.38^d	2.29^d	0.594^d	3.79^d	12.2^c	26.1^d
	L-VT09	2.78^a	3.89^c	0.900^{ab}	3.41^c	2.34^c	0.692^c	4.60^c	16.5^b	36.1^c
	N-VT06	2.32^c	4.25^b	0.870^b	3.60^b	4.02^b	0.796^b	5.76^b	16.2^b	37.8^b
	L-VT09 + N-VT06	2.58^b	5.61^a	0.937^a	4.64^a	5.34^a	1.024^a	6.44^a	18.8^a	45.3^a
P (A)		*	*	*	*	*	*	*	*	*
P (B)		*	*	*	*	*	*	*	*	*
P (A×B)		ns	*	*	*	*	*	*	*	*
CV, %		7.54	10.7	9.06	7.11	5.38	7.72	11.7	6.85	3.75

From the result of interaction analysis, at 100% P fertilizer level, not supplying bacteria had lower total P uptake than supplying the mixture of the *B. silvatlantica* L-VT09 and N-VT06 strains, or only the L-VT09 strain or only the N-VT06 strain, with 32.6 compared with 56.0, 42.1, and 47.5 kg ha^{-1}, respectively. At 50% P, supplying only the L-VT09 strain and only the N-VT06 strain had equivalent total P uptakes to that in the treatment fertilized with 100% P without bacteria, which were 33.7, 34.5, and 32.6 kg ha^{-1}, respectively. In addition, the treatment supplied with the mixture of the L-VT09 and N-VT06 strains plus 25% P resulted in higher total P uptake than the treatment fertilized with 100% P according to recommendations, with 34.9 compared with 32.6 kg ha^{-1}, respectively (Figure 1).

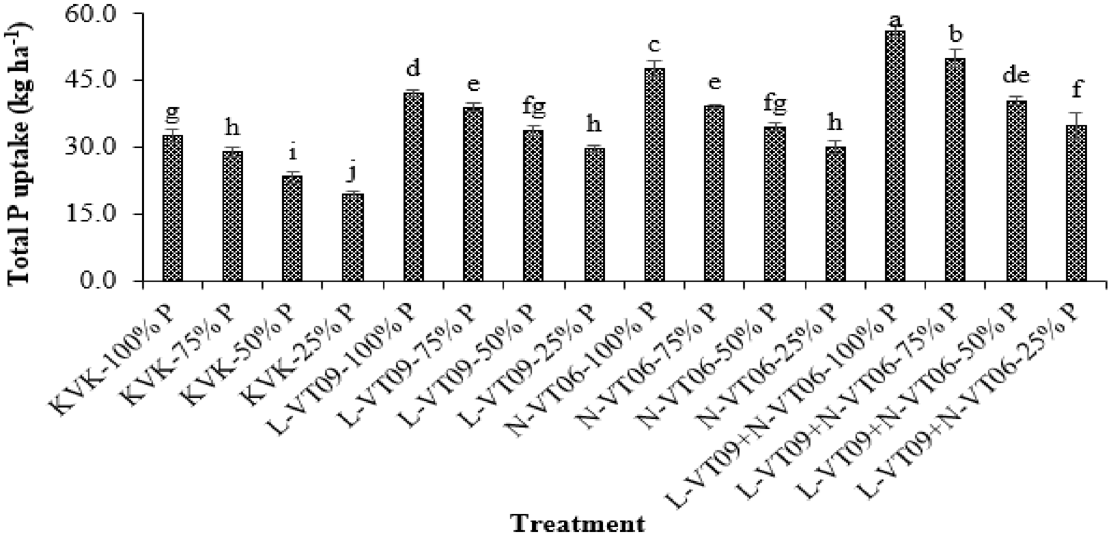

Figure 1. Effects of P-solubilizing bacterial endophytes, *Burkholderia silvatlantica* L-VT09 and N-VT06, on total P uptake of pineapple in acid sulfate soil. KVK: No bacteria.

Effects on growth, yield components, and yield of pineapples in acid sulfate soil

Growth of pineapple in the acid sulfate soil. Table 6 reveals that the treatment fertilized with 100% P resulted in higher plant height, length, and width of D-leaf, length, and diameter of stem, length, and diameter of peduncle, and length and diameter of crown than the treatment fertilized with 25% P did, with 77.0, 63.6, 6.31, 16.3, 4.83, 24.2, 3.23, 17.2, and 4.79 cm compared with 71.9, 59.3, 5.76, 14.1, 4.48, 21.3, 2.78, 12.1, and 4.06 cm, respectively.

Lengths of D-leaf remained statistically between treatments supplied with either only the *B. silvatlantica* L-VT09 or the *B. silvatlantica* N-VT06 strain and the treatment without bacteria (60.4, 61.2, and 60.1 cm, respectively), which were all lower than in the treatment supplied with the mixture of the L-VT09 and N-VT06 strains (62.9 cm). For lengths of the stem, the treatments supplied with only the N-VT06 strain and the one supplied with the mixture of the L-VT09 and N-VT06 strains were not significantly different from each other and all longer than those in the treatment with only the L-VT09 strain and in the one without bacteria supplied, valuing at 16.1 ~ 16.7 > 14.2 ~ 13.3 cm, respectively. For lengths of peduncle, the treatment supplied with the mixture of the L-VT09 and N-VT06 strains differed not significantly from the treatment supplied with only the L-VT09 strain (24.0 and 23.7 cm) and longer than those in the treatment supplied with only the N-VT06 and in the one without bacteria (22.1 and 21.6 cm) (Table 6).

The treatments supplied with the mixture of the *B. silvatlantica* L-VT09 and N-VT06 strains, with only the L-VT09 or N-VT06 strain, had plant height, D-leaf width, stem diameter, peduncle diameter, and crown length and width in ranges of 75.2-77.7, 6.10-6.14, 4.58-4.87, 2.93-3.35, 15.4-18.8, and 4.65-4.88 cm, which all outweighed those in the treatment without bacteria, where results were 69.4, 5.79, 4.43, 2.74, 10.2, and 3.89 cm, respectively. For D-leaf length, only in the treatment supplied with the mixture of the L-VT09 and N-VT06, the result was higher than in the treatment without bacteria, with 62.9 compared with 60.1 cm. However, stem lengths in the treatments supplied with the mixture of the L-VT09 and N-VT06 strains and with only the N-VT06 strain (23.7-24.0 cm) were longer than those in the treatment without bacteria (21.6 cm) (Table 6).

Table 6. Effects of P-solubilizing bacterial endophytes, *Burkholderia silvatlantica* L-VT09 and N-VT06, on the growth of pineapple in acid sulfate soil. Numbers followed by different letters are significantly different. *Different at 5% significance; ^{ns}: nonsignificant; A: fertilizer factor; B: bacteria factor; L-VT09: supplying only the P-solubilizing *B. silvatlantica* L-VT09 strain; N-VT06: supplying only the P-solubilizing *B. silvatlantica* N-VT06; L-VT09 + N-VT06: supplying the mixture of P-solubilizing *B. silvatlantica* L-VT09 and N-VT06.

Factors		Plant height	Leaves per plant	D-leaf length	D-leaf width	Stem length	Stem diameter	Peduncle length	Peduncle diameter	Slips number per plant	Crown length	Crown diameter
		cm	leaves plant^{-1}	cm						slips plant^{-1}	cm	
Level of P fertilizer (A)	100% P	77.0^{a}	50.7^{a}	63.6^{a}	6.31^{a}	16.3^{a}	4.83^{a}	24.2^{a}	3.23^{a}	9.29^{a}	17.2^{a}	4.79^{ab}
	75% P	75.1^{ab}	51.8^{a}	61.8^{b}	6.18^{a}	15.3^{ab}	4.68^{b}	23.3^{b}	3.05^{ab}	8.95^{ab}	15.9^{b}	4.86^{a}
	50% P	73.9^{bc}	50.7^{a}	60.1^{c}	5.88^{b}	14.7^{b}	4.60^{bc}	22.6^{c}	2.93^{bc}	8.44^{bc}	15.0^{b}	4.49^{b}
	25% P	71.9^{c}	47.9^{b}	59.3^{c}	5.76^{b}	14.1^{b}	4.48^{c}	21.3^{d}	2.78^{c}	8.04^{c}	12.1^{c}	4.06^{c}
Supplementation of bacteria (B)	No bacteria	69.4^{c}	46.2^{c}	60.1^{b}	5.79^{b}	13.3^{b}	4.43^{c}	21.6^{b}	2.74^{c}	8.31	10.2^{c}	3.89^{b}
	L-VT09	75.2^{b}	50.1^{b}	60.4^{b}	6.10^{a}	14.2^{b}	4.71^{b}	23.7^{a}	2.96^{b}	8.63	15.4^{b}	4.82^{a}
	N-VT06	75.6^{ab}	52.0^{ab}	61.2^{b}	6.14^{a}	16.1^{a}	4.58^{b}	22.1^{b}	2.93^{b}	8.63	15.7^{b}	4.65^{a}
	L-VT09 + N-VT06	77.7^{a}	52.8^{a}	62.9^{a}	6.11^{a}	16.7^{a}	4.87^{a}	24.0^{a}	3.35^{a}	9.16	18.8^{a}	4.84^{a}
P (A)		*	*	*	*	*	*	*	*	*	*	*
P (B)		*	*	*	*	*	*	*	*	ns	*	*
P (A×B)		ns	*	*	ns	ns	ns	ns	ns	ns	*	ns
CV, %		3.96	5.78	2.96	3.90	11.4	4.19	3.70	8.56	12.8	8.88	9.41

The treatments fertilized with P fertilizer from 50% to 100% P reached a higher number of leaves per plant than the treatment fertilized with 25% P, with 50.7-51.8 compared with 47.9 leaves plant^{-1}. In addition, supplying the single strain and the mixed strains both enhanced the number of leaves. The treatments fertilized with 75% and 100% P had higher slips number than the treatment fertilized with 25% P, with 8.95-9.29 compared with 8.04 slips plant^{-1}. All the treatments supplied bacteria were equivalent in the number of slips per plant, ranging from 8.31 to 9.16 slips plant^{-1} (Table 6).

Yield components and yield of pineapples in acid sulfate soil. Table 7 indicates that the fruit length and fruit diameter changed between levels of P fertilizer and bacteria supplementations ($P < 0.05$). In detail, the treatment fertilized with 100% P reached the longest and widest fruit (18.5 and 9.10 cm), while the smallest result was in the treatment fertilized with 25% P (15.0 and 7.80 cm). Fruit lengths in the treatment supplied with only the *B. silvatlantica* N-VT06 strain and in the treatment supplied with the mixture of *B. silvatlantica* L-VT09 and N-VT06 strains were statistically the same (18.3-18.4 cm); then, the second longest fruit was in the treatment supplied with only the L-VT09 strain (16.0 cm) and the shortest one was in the treatment without bacteria, 14.8 cm. Fruit diameter peaked in the treatment supplied with the mixture of the L-VT09 and N-VT06 strains and bottomed without bacteria, with 9.12 and 7.66 cm, respectively.

Table 7. Effects of P-solubilizing bacterial endophytes, *Burkholderia silvatlantica* L-VT09 and N-VT06, on yield components and yield of pineapple in acid sulfate soil. Numbers followed by different letters are significantly different. *Different at 5% significance; ns: nonsignificant; A: fertilizer factor; B: bacteria factor; L-VT09: supplying only the P-solubilizing *B. silvatlantica* L-VT09 strain; N-VT06: supplying only the P-solubilizing *B. silvatlantica* N-VT06; L-VT09 + N-VT06: supplying the mixture of P-solubilizing *B. silvatlantica* L-VT09 and N-VT06.

Factors		Fruit length	Fruit diameter	One-fruit weight	Yield
		cm	cm	kg	t ha^{-1}
Level of P fertilizer (A)	100% P	18.5^{a}	9.10^{a}	1.238^{a}	35.8^{a}
	75% P	17.6^{b}	8.60^{b}	1.156^{a}	33.1^{b}
	50% P	16.3^{c}	8.40^{b}	0.955^{b}	27.7^{c}
	25% P	15.0^{d}	7.80^{c}	0.789^{c}	23.7^{d}
Supplementation of bacteria (B)	No bacteria	14.8^{c}	7.66^{c}	0.833^{c}	24.0^{d}
	L-VT09	16.0^{b}	8.54^{b}	0.997^{b}	28.7^{c}
	N-VT06	18.4^{a}	8.58^{b}	1.066^{b}	32.1^{b}
	L-VT09 + N-VT06	18.3^{a}	9.12^{a}	1.243^{a}	35.5^{a}
P (A)		*	*	*	*
P (B)		*	*	*	*
P (A×B)		*	ns	*	*
CV, %		5.54	4.35	11.2	5.66

The treatments fertilized with 100% and 75% P had equivalent weights per fruit each other and were higher than those in the treatments fertilized with 50% and 25% P, with 1.238 and 1.156 compared to 0.955 and 0.789 kg, respectively. For the factor of bacteria supplementations, the treatment without bacteria had the lightest fruit weight, with 0.833 kg, and the heaviest one was in the treatment supplied with the mixture of the *B. silvatlantica* L-VT09 and N-VT06 strains, with 1.24 kg (Table 7).

The yield gradually dropped when levels of P fertilizer reduced, with 100%, 75%, 50%, and 25% P corresponding to 35.8 > 33.1 > 27.7 > 23.7 t ha^{-1}. Moreover, the treatments supplied with the mixture of the L-VT09 and N-VT06 strains, with only the N-VT06 strain, with only the L-VT09 strain had higher yield than the treatment without bacteria did, with 28.7-35.5 compared with 24.0 t ha^{-1} (Table 7).

From the analysis of interactions, the yield in the treatments supplied with only the *B. silvatlantica* L-VT09 strain (34.4 t ha^{-1}), with only the *B. silvatlantica* N-VT06 strain (34.8 t ha^{-1}) and the mixture of the L-VT09 and N-VT06 (43.1 t ha^{-1}) was higher than in the treatment without bacteria (30.9 t ha^{-1}) when 100% P based on the recommendation was applied. Yield values at 100% P without bacteria were equivalent to those in the treatment supplied with only the N-VT06 strain combined with 75% P (33.5 t ha^{-1}) and lower than those in the treatments fertilized with 75% P plus either the single strain of L-VT09 or the mixture of the L-VT09 and N-VT06 strains (34.3 and 36.2 t ha^{-1}). The treatment supplied with the mixture of the L-VT09 and N-VT06 strains combined with either 50% or 25% P had a statistically equal yield to that in the treatment fully fertilized according to the recommendation, whose yield was 30.9-31.8 t ha^{-1}. At the same level of P fertilizer (25% P), combining with the mixture of the L-VT09 and N-VT06 strains resulted in higher than those in the treatments without bacteria and with only the L-VT09 strain, i.e., 31.1 compared to 14.0 and 20.1 t ha^{-1}, respectively. Furthermore, supplying the mixture of the L-VT09 and N-VT06 strains plus 50% or 25% P (31.8 and 31.1 t ha^{-1}) remained, in comparison to supplying only the *B. silvatlantica* N-VT06 strain plus 50% and 25% P (30.5 and 29.7 t ha^{-1}) (Figure 2).

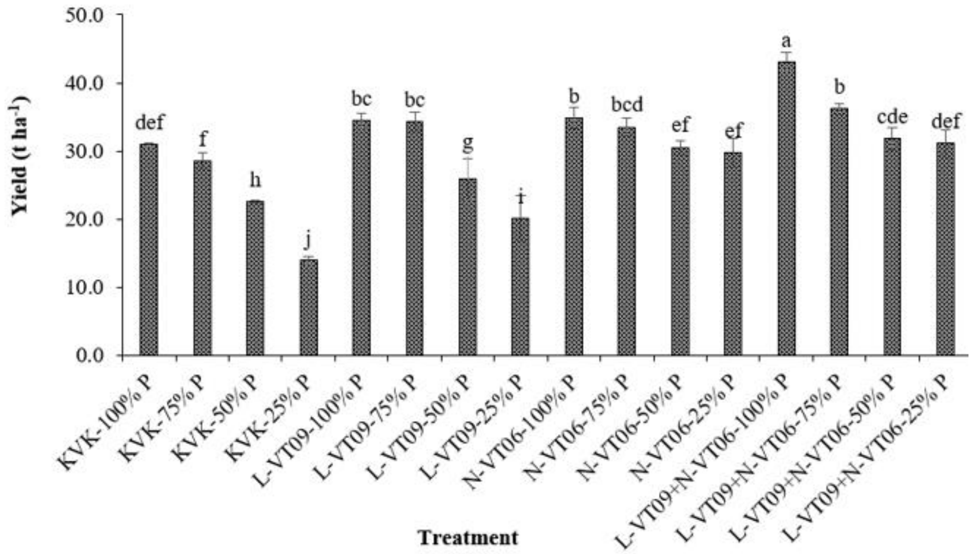

Figure 2. Effects of P-solubilizing bacterial endophytes, *Burkholderia silvatlantica* L-VT09 and N-VT06, on the yield of pineapple in acid sulfate soil. KVK: No bacteria.

Effects on the fruit quality of pineapples in acid sulfate soil

Table 8 shows insignificantly different values of pH, vitamin C content, and total acid between levels of P fertilizer and supplementations of bacteria. In particular, these values fluctuated at 3.53-3.63, 13.1-14.0 mg 100 g^{-1}, and 21.3-23.1 mg L^{-1} between levels of P fertilizer, 3.53-3.63, 11.9-14.6 mg 100 g^{-1} and 21.3-22.7 mg L^{-1} between supplementations of bacteria, respectively.

Table 8. Effects of P-solubilizing bacterial endophytes, *Burkholderia silvatlantica* L-VT09 and N-VT06, on fruit quality of pineapple in acid sulfate soil. Numbers followed by different letters are significantly different. *Different at 5% significance; ns: nonsignificant; A: fertilizer factor; B: bacteria factor; L-VT09: supplying only the P-solubilizing *B. silvatlantica* L-VT09 strain; N-VT06: supplying only the P-solubilizing *B. silvatlantica* N-VT06; L-VT09 + N-VT06: supplying the mixture of P-solubilizing *B. silvatlantica* L-VT09 and N-VT06.

Factors		pH	Sugar content	Water content	Vitamin C content	Acid total content	Fruit color		
			°Brix	mL	mg 100 g^{-1}	mg L^{-1}	L*	a*	b*
Level of P fertilizer (A)	100% P	3.57	8.79	362.4^{a}	13.1	22.1	202.3^{a}	42.7^{ab}	86.9^{b}
	75% P	3.53	9.08	257.2^{b}	13.3	21.4	198.5^{ab}	43.8^{a}	90.4^{ab}
	50% P	3.56	9.11	211.6^{c}	14.0	23.1	194.3^{bc}	40.4^{b}	91.8^{ab}
	25% P	3.63	9.17	175.1^{d}	13.4	21.3	188.4^{c}	35.9^{c}	94.5^{a}
Supplementation of bacteria (B)	No bacteria	3.63	8.14^{c}	191.4^{c}	12.8	21.3	174.1^{c}	31.6^{c}	106.8^{a}
	L-VT09	3.59	8.81^{b}	197.2^{c}	14.6	22.3	198.6^{b}	42.5^{b}	84.8^{b}
	N-VT06	3.53	9.36^{ab}	260.0^{b}	14.4	22.7	210.7^{a}	45.6^{a}	84.9^{b}
	L-VT09 + N-VT06	3.55	9.85^{a}	357.8^{a}	11.9	21.6	200.2^{b}	43.2^{ab}	87.1^{b}
P (A)		ns	ns	*	ns	ns	*	*	*
P (B)		ns	*	*	ns	ns	*	*	*
P (A×B)		ns	ns	*	ns	ns	*	*	ns
CV, %		3.30	9.13	9.42	23.8	15.1	4.84	9.92	7.75

Sugar content was insignificantly different between P fertilizer levels but significantly different between supplementations of bacteria (Table 8). For the P fertilizer application, the sugar content was from 8.79 to 9.17 °Brix. When supplying the mixture of the *B. silvatlantica* L-VT09 and N-VT06 strains (9.85 °Brix), the sugar content was higher than that for supplying with only the L-VT09 strain (8.81 °Brix). However, all individual and mixture supplementations of bacteria resulted in higher sugar content than those in the treatment without bacteria (8.14 °Brix).

Water contents went down when levels of P fertilizer dropped, with 362.4 > 257.2 > 211.6 > 175.1 mL corresponding to 100%, 75%, 50%, and 25% P. The treatment supplied with the mixture of the L-VT09 and N-VT06 strains had higher water content than that in the treatments with only either the L-VT09 or the N-VT06 strain and the one without bacteria, with 357.8 > 260.0 > 197.2 ~ 191.4 mL, respectively. To be more specific, the treatment with only the L-VT09 strain had equivalent contents of water compared to that in the treatment without bacteria (Table 8).

Table 8 reveals that the treatments fertilized with 100% and 75% P resulted in equivalent values of L* and a* and higher than those in the treatment fertilized with 25% P, with 202.3 ~ 198.5 compared to 188.4 and 42.7 ~ 43.8 compared to 35.9, respectively. Values of b* varied insignificantly between treatments fertilized with 75%, 50%, and 25% P (90.4, 91.8, and 94.5, sequentially). However, the treatment fertilized with 25% P had higher b* values than those in the treatment fertilized with 100% P (86.9). For the treatments supplied bacteria, L* values peaked in the treatment supplied with only the *B. silvatlantica* N-VT06 strain (210.7) and reached the bottom in the treatment without bacteria (174.1). Values of a* in the treatments supplied with bacteria (42.5-45.6) were higher than those in the treatment without ones (31.6). The treatments supplied with only the *B. silvatlantica* L-VT09 strain, with only the N-VT06 strain, and with the mixture of the L-VT09 and N-VT06 strains possessed b* values equivalent to each other and lower than that in the treatment without bacteria, with 84.8-87.1 compared to 106.8.

DISCUSSION

Phosphorus is a well-known macronutrient that is essential for plant growth and development (Johan et al., 2021). From the analytic results in Table 1, the soil for the experiment had a low pH value, leading to low availability of nutrients, per the study by Oshunsanya (2018). In addition, pineapple can adapt well to a pH of 4.5-5.5 (Vásquez-Jiménez and Bartholomew, 2018). However, Johan et al. (2021) assumed that P immobilization depends on pH and oxides, e.g., Fe, Al, and Ca, accounting the most in the acid sulfate soil, which reduces the P availability due to precipitation between P and Fe, Al, and Ca. The result revealed that increasing levels of P fertilizer led to an increase in available P content and concentrations of Fe-P, Al-P, and Ca-P, while supplying the P-solubilizing bacterial endophytes *B. silvatlantica* L-VT09 and *B. silvatlantica* N-VT06 contributed to enhancing available P content and declining concentrations of Fe-P, Al-P, and Ca-P, compared with those in the treatment without bacteria (Table 2). Thereby, this biological approach for solubilizing P is promising. In particular, P-solubilizing bacteria play an important role in ameliorating soil fertility (Etesami and Jeong, 2021), enhancing solubilization of insoluble P, improving secretions of phosphatase acid enzyme and contents of available P, and ultimately promoting plant growth (Wang et al., 2017; Song et al., 2022). Moreover, Elhaissoufi et al. (2021) also stated that P-solubilizing bacteria increase available P content and boost P use efficiencies, improving plant development. In addition, supplying the P-solubilizing bacterial endophytes, L-VT09 and N-VT06, raised the concentrations of NH_4^+ and NO_3^- in the soil compared to when no bacteria were supplied; the enhancement was by 5.30-15.5 mg NH_4^+ kg^{-1} and 0.83-1.22 mg NO_3^- kg^{-1}. Furthermore, bacterial densities in the treatment supplied with the mixture of L-VT09 and N-VT06 strains were higher than that in the treatment without bacteria (Table 2). Levels of P fertilizer and supplementations of bacteria had not affected values of pH_{H2O}, pH_{KCl}, EC, total N and total P in soil yet (Table 2). According to Kartikawati et al. (2020), the bacterial endophytes *Pseudomonas* sp., *Brevibacillus* sp., and *Mesorhizobium* sp. are able to improve the chemical characteristics of soil for pepper. Two strains of bacterial endophytes, L-VT09 and N-VT06 strains, have been studied and proven to solubilize Fe-P, Al-P, and Ca-P by Khuong et al. (2022). In addition, Divjot et al. (2021) also reported that bacterial endophytes belonging to *Burkholderia* sp. are efficient in solubilizing P.

Fertilization with an increasing amount of P fertilizer enhanced dry biomass in stovers of plants, leading to high concentrations of P when the amount of P fertilizer applied increased. Furthermore, supplying P-solubilizing bacterial endophytes improved dry biomass, P uptakes, and P concentrations in plant stovers (Tables 3 and 4). The result of interaction analysis showed that supplying bacteria plus 100% and 75% P led to higher P uptake than those in the treatment fertilized with 100% P. Significantly, fertilizing 25% P plus supplying the mixture of L-VT09 and N-VT06 strains had higher total P uptake than that in the treatment fertilized with 100% P did, with a difference of 2.30 kg ha^{-1} (Figure 1). These results show that the P-solubilizing bacterial endophytes have improved nutrient absorption and accumulation in plants and ameliorated available soil nutrients, especially the P nutrient. This is because the P-solubilizing bacteria can produce organic acids, such as gluconic acid, oxalic acid, tartaric acid, and lactic acid, facilitating P-solubilization of inorganic compounds (Billah et al., 2019). Furthermore, for organic P compounds, enzymatic activities of phosphatase, phytase, and C–P lyase are the key mechanisms (Othman and Panhwar, 2014). This was in accordance with previous studies. Bacterial endophytes help increase plant biomass and improve P uptake for the plant (Divjot et al., 2021).

The P-solubilizing bacteria contribute to plant growth and improve P availability, producing a better plant yield (Divjot et al., 2021). The result revealed that characteristics of growth, yield components, and fruit yield of pineapples corresponded to a reduction in P fertilizer levels. Moreover, the treatment supplied with bacteria had superior growth parameters to those without bacteria (Table 6). Furthermore, yield components, such as fruit length, fruit diameter, and weight per fruit, in the treatments supplied with bacteria were higher than in the treatment without bacteria. The treatments supplied with the bacteria reached a higher yield than those without bacteria (Table 7). The interaction analysis result showed that at the same 100% P according to the recommendation, yield in the treatment without bacteria was lower than those in the treatments supplied with the mixture *B. silvatlantica* L-VT09 and N-VT06, with only the L-VT09 strain and with only the N-VT06 strain (Figure 2). This was in accordance with the study by Kartikawati et al. (2020), where supplying the mixture of bacterial endophytes *Pseudomonas* sp., *Brevibacillus* sp., and *Mesorhizobium* sp. promotes growth and yield in black pepper. Applying P-solubilizing bacteria, *Bacillus mojavensis* and *B. megaterium*, improves yield components and yield in sugarcane (Chungopast et al., 2021). Besides, a genus of bacterial endophytes, *Burkholderia* sp., stimulates growth (Mannaa et al., 2018) and yield of cultivars (Divjot et al., 2021). The improvement of growth and yield of the plants can be attributed to not only the amelioration of soil fertility, as mentioned previously, but also the ability to produce plant growth-promoting substances, such as IAA and siderophores by the *B. silvatlantica* species (Alves et al., 2016).

Contents of vitamin C, total acid, and pH had not been influenced by changes in P fertilizer levels and bacteria supplementations. Spironello et al. (2004) also reported that vitamin C content does not change by different P fertilizer levels. However, sugar content, water content, and fruit color were changed by treatments supplied with bacteria. The treatment supplied with bacteria had higher sugar content and water content than the treatment without bacteria. This was consistent with the study by Huu et al. (2022). Moreover, in the treatment supplied with bacteria, values of L* and a* were higher than those in the treatment without bacteria, while b* values followed an opposite trend (Table 8). In the meantime, a^* and b^* values present mature levels of fruits, and L* shows the darkness and lightness of the color (Itle and Kabelka, 2009); that is, high L* presents the bright and fresh fruits, low a^* and b^* represent green fruits, and high a^* and b^* indicate changes in colors of fruit shell due to chlorophyll reduction and synthesis of carotenoid (Christ and Hörtensteiner, 2014). In the current study, the * and b* values were opposite, so the fruits were not too green or too ripening, and they were brighter. In other words, with the application of the bacteria mixture and the P fertilizer, fruits looked fresher and appeared better than the others. Ultimately, the bacteria improved the fruit's quality in colors and chemical components.

CONCLUSIONS

The current study has shown that supplying with the mixture of *Burkholderia silvatlantica* L-VT09 and N-VT06 strains resulted in better available P concentration, bacterial densities, plants growth, yield, and fruit quality of pineapples in comparison with those in the treatment without bacteria. Simultaneously, supplying with the mixture of the *B. silvatlantica* L-VT09 and N-VT06 strains had lower concentrations of Al-P, Fe-P, and Ca-P and fruit colors (b*) than those in the treatment without bacteria. Supplying the mixture of *B. silvatlantica* L-VT09 and N-VT06 strains combined with 25% P according to recommendation still maintained the pineapple yield in the acid sulfate soil. Therefore, the bacteria have been proven to be a good biofertilizer and bioremediator that enhanced pineapple performance in acid sulfate soil and the soil fertility. Furthermore, the biofertilizer should be tested on a large scale. Moreover, other crops should be applied with these bacteria. Ultimately, the biofertilizer can be commercialized and transferred for farmers' use.

Author contribution

Conceptualization: N.Q.K., N.T.P. Methodology: N.Q.K., N.T.P. Formal analysis: N.Q.K., N.V.T., L.N.T.X., L.T.M.T., L.T.Q. Investigation: N.Q.K, N.V.T., L.N.T.X., L.T.M.T., L.T.Q. Data curation, writing the original draft, project administration, and funding acquisition: N.Q.K. Writing-review & editing: L.T.Q., N.T.P. Supervision: N.T.P. All co-authors reviewed the final version and approved the manuscript before submission.

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References

Alves, G.C., de Matos Macedo, A.V., Dos Reis, F.B., Urquiaga, S., Reis, V.M. 2016. Plant growth promotion by four species of the genus *Burkhoderia*. Plant and Soil 399:373-387. doi:10.1007/s11104-015-2701-4.

Be, L.V., Hoa, L.V. 2009. Comparing growth and fruit weight of Queen pineapple between sucker material. Can Tho University Journal of Science 11:159-167.

Billah, M., Khan, M., Bano, A., Hassan, T.U., Munir, A., Gurmani, A.R. 2019. Phosphorus and phosphate solubilizing bacteria: Keys for sustainable agriculture. Geomicrobiology Journal 36(10):904-916. doi:10.1080/01490451.2019.1654043.

Chandran, H., Meena, M., Swapnil, P. 2021. Plant growth-promoting rhizobacteria as a green alternative for sustainable agriculture. Sustainability 13(19):10986. doi:10.3390/su131910986.

Christ, B., Hörtensteiner, S. 2014. Mechanism and significance of chlorophyll breakdown. Journal of Plant Growth Regulation 33:4-20. doi:10.1007/s00344-013-9392-y.

Chungopast, S., Thongjoo, C., Islam, A.K.M., Yeasmin, S. 2021. Efficiency of phosphate-solubilizing bacteria to address phosphorus fixation in Takhli soil series: A case of sugarcane cultivation, Thailand. Plant and Soil 460:347-357. doi:10.1007/s11104-020-04812-w.

da Silva, J.R., Netto, A.T., de Medeiros, B.P., de Deus, B.C.D.S., Silva, M.V.S., Ferraz, T.M., et al. 2020. Endophytic diazotrophic bacteria mitigate water deprivation effects in pineapple explants during acclimatization. Theoretical and Experimental Plant Physiology 32:63-77. doi:10.1007/s40626-020-00168-9.

Dhankhar, R., Sheoran, S., Dhaka, A., Soni, R. 2013. The role of phosphorus solubilizing bacteria (PSB) in soil management-an overview. International Journal of Development Research 3(9):31-36.

Divjot, K.O.U.R., Rana, K.L., Tanvir, K.A.U.R., Yadav, N., Yadav, A.N., Kumar, M., et al. 2021. Biodiversity, current developments and potential biotechnological applications of phosphorus-solubilizing and-mobilizing microbes: A review. Pedosphere 31(1):43-75. doi:10.1016/S1002-0160(20)60057-1.

Elhaissoufi, W., Ghoulam, C., Barakat, A., Zeroual, Y., Bargaz, A. 2021. Phosphate bacterial solubilization: a key rhizosphere driving force enabling higher P use efficiency and crop productivity. Journal of Advanced Research 38:13-28. doi:10.1016/j.jare.2021.08.014.

Etesami, H. 2020. Enhanced phosphorus fertilizer use efficiency with microorganisms. p. 215-245. In Meena, R. (ed.) Nutrient dynamics for sustainable crop production. Springer, Singapore. doi:10.1007/978-981-13-8660-2_8.

Etesami, H., Jeong, B.R. 2021. Contribution of arbuscular mycorrhizal fungi, phosphate-solubilizing bacteria, and silicon to P uptake by plant: A review. Frontiers in Plant Science 12:699618. doi:10.3389/fpls.2021.699618.

Flatian, A.N., Anas, I., Sutandi, A. 2021. The ability of some microbes to solubilize the hardly soluble phosphorous and potassium from various sources in vitro. In IOP Conference Series: Earth and Environmental Science. 648:012143. doi:10.1088/1755-1315/648/1/012143.

Guerrant, G.O., Lambert, M.A., Moss, C.W. 1982. Analysis of short-chain acids from anaerobic bacteria by high-performance liquid chromatography. Journal of Clinical Microbiology 16(2):355-360. doi:10.1128/jcm.16.2.355-360.1982.

Houba, V.J.G., Van der Lee, J.J., Novozamsky, I., Walinga, I. 1988. Soil and plant analysis. Part 5: Soil analysis procedures. Department of Soil Science and Plant Nutrition, Wageningen Agricultural University, The Netherlands.

Huu, T.N., Giau, T.T.N., Ngan, P.N., Van, T.T.B., Khuong, N.Q. 2022. Potential of phosphorus solubilizing purple nonsulfur bacteria isolated from acid sulfate soil in improving soil property, nutrient uptake, and yield of pineapple (*Ananas comosus* L. Merrill) under acidic stress. Applied and Environmental Soil Science 2022:8693479. doi:10.1155/2022/8693479.

Itle, R.A., Kabelka, E. A. 2009. Correlation between L* a* b* color space values and carotenoid content in pumpkins and squash (*Cucurbita* spp.) HortScience 44(3):633-637. doi:10.21273/HORTSCI.44.3.633.

Johan, P.D., Ahmed, O.H., Omar, L., Hasbullah, N.A. 2021. Phosphorus transformation in soils following co-application of charcoal and wood ash. Agronomy 11(10):2010. doi:10.3390/agronomy11102010.

Kartikawati, A., Nurhayati, H., Sarwendah, M. 2020. Plant growth promoting effects of indigenous rhizosphere and endophytic bacteria on soil fertility and black pepper (*Piper nigrum*) yield at Bangka Belitung. IOP Conference Series: Earth and Environmental Science 418:012057. doi:10.1088/1755-1315/418/1/012057.

Kim, J.H., Kim, S.J., Nam, I.H. 2021. Effect of treating acid sulfate soils with phosphate solubilizing bacteria on germination and growth of tomato (*Lycopersicon esculentum* L.) International Journal of Environmental Research and Public Health 18(17):8919. doi:10.3390/ijerph18178919.

Khuong, N.Q., Tuong, N.V., Truong, C.M., Qui, C.N., Nhan, T.C., Xuan, L.N.T., et al. 2022. Selection and identification of phosphate solubilizing endophytic bacteria from pineapple cultivated on acid sulfate soil in Vi Thanh city, Hau Giang province. Hue University of Agriculture and Forestry Journal of Agricultural Science and Technology 6(3):3169-3179. doi:10.46826/huaf-jasat.v6n3y2022.948.

Mannaa, M., Park, I., Seo, Y.S. 2018. Genomic features and insights into the taxonomy, virulence, and benevolence of plant-associated *Burkholderia* species. International Journal of Molecular Sciences 20(1):121. doi:10.3390/ijms20010121.

Naoki, H., Nishiyama, M., Matsumoto, S. 2001. Inhibition of methanogens increases photo-dependent nitrogenase activities in anoxic paddy soil amended with rice straw. FEMS Microbiology Ecology 35(3):231-238. doi:10.1111/j.1574-6941.2001.tb00808.x.

Oshunsanya, S.O. 2018. Introductory chapter: Relevance of soil pH to agriculture. p. 1-4. In Oshunsanya, S.O. (ed.) Soil pH for nutrient availability and crop performance. IntechOpen. doi:10.5772/intechopen.82551.

Othman, R., Panhwar, Q.A. 2014. Phosphate-solubilizing bacteria improves nutrient uptake in aerobic rice. In Khan, M., Zaidi, A., Musarrat, J. (eds.) Phosphate solubilizing microorganisms. Springer, Cham, Switzerland. doi:10.1007/978-3-319-08216-5_9.

Percival, L.M.E., Bond, D.P.G., Rakociński, M., Marynowski, L., Hood, A.V.S., Adatte, T., et al. 2020. Phosphorus-cycle disturbances during the Late Devonian anoxic events. Global and Planetary Change 184:103070. doi:10.1016/j.gloplacha.2019.103070.

Rafi, M.M., Krishnaveni, M.S., Charyulu, P.B.B.N. 2019. Phosphate-solubilizing microorganisms and their emerging role in sustainable agriculture. p. 223-233. In Buddolla, V. (ed.) Recent developments in applied microbiology and biochemistry. Academic Press, Cambridge, Massachusetts, USA. doi:10.1016/B978-0-12-816328-3.00017-9.

Rai, P.K., Singh, M., Anand, K., Saurabh, S., Kaur, T., Kour, D., et al. 2020. Role and potential applications of plant growth-promoting rhizobacteria for sustainable agriculture. p. 49-60. In Rastegari, A.A., Yadav, A.N., Yadav, N. (eds.) New and future developments in microbial biotechnology and bioengineering. Elsevier, Amsterdam, The Netherlands. doi:10.1016/B978-0-12-820526-6.00004-X.

Rana, K.L., Kour, D., Kaur, T., Devi, R., Yadav, A., Yadav, A.N. 2021. Bioprospecting of endophytic bacteria from the Indian Himalayas and their role in plant growth promotion of maize (*Zea mays* L.) Journal of Applied Biology & Biotechnology 9(3):41-50. doi:10.7324/JABB.2021.9306.

Saini, R.S., Sharma, K.D., Dhankhar, O.P., Kaushik, R.A. 2001. Laboratory manual of analytical techniques in horticulture. Agrobios, Jodhpur, India.

Santoyo, G., Moreno-Hagelsieb, G., del Carmen Orozco-Mosqueda, M., Glick, B.R. 2016. Plant growth-promoting bacterial endophytes. Microbiological Research 183:92-99. doi:10.1016/j.micres.2015.11.008.

Song, C., Wang, W., Gan, Y., Wang, L., Chang, X., Wang, Y., et al. 2022. Growth promotion ability of phosphate-solubilizing bacteria from the soybean rhizosphere under maize-soybean intercropping systems. Journal of the Science of Food and Agriculture 102(4):1430-1442. doi:10.1002/jsfa.11477.

Sparks, D.L., Page, A.L., Helmke, P.A., Loeppert, R.H., Soltanpour, P.N., Tabatabai, M.A., et al. (eds.) 1996. Methods of soil analysis. Part 3-Chemical methods. SSSA Book Series 5.3. SSSA, ASA, Madison, Wisconsin, USA. doi:10.2136/sssabookser5.3.

Spironello, A., Quaggio, J.A., Teixeira, L.A.J., Furlani, P.R., Sigrist, J.M.M. 2004. Pineapple yield and fruit quality effected by NPK fertilization in a tropical soil. Revista Brasileira de Fruticultura 26(1):155-159.

Vásquez-Jiménez, J., Bartholomew, D.P. 2018. Plant nutrition. p. 175-202. In Sanewski, G.M., Bartholomew, D.P., Paull, R.E. (eds.) The pineapple: Botany, production and uses. CABI Publishing, Wallingford, UK. doi:10.1079/9781786393302.0175.

Wang, Z., Xu, G., Ma, P., Lin, Y., Yang, X., Cao, C. 2017. Isolation and characterization of a phosphorus-solubilizing bacterium from rhizosphere soils and its colonization of Chinese cabbage (*Brassica campestris* ssp. *chinensis*). Frontiers in Microbiology 8:1270. doi:10.3389/fmicb.2017.01270.

Yadav, B.K., Verma, A. 2012. Phosphate solubilization and mobilization in soil through microorganisms under arid ecosystems. p. 93-108. In Ali M. (eds.) The functioning of ecosystems. IntechOpen, Rijeka, Croatia. doi:10.5772/35917.