Journal of Advances in Microbiology Research

E-ISSN: 2709-944X
P-ISSN: 2709-9431
JRM 2024; 5(2): 145-150

www.microbiojournal.com
Received: 21-07-2024
Accepted: 24-08-2024

Douglas
Department of Microbiology, Faculty of Science, Rivers State University, PMB 5080, Nkpolu-Oroworukwo, Port Harcourt, Rivers State, Nigeria

Salome Ibietela
Department of Microbiology, Faculty of Science, Rivers State University, PMB 5080, Nkpolu-Oroworukwo, Port Harcourt, Rivers State, Nigeria

Nrior
Department of Microbiology, Faculty of Science, Rivers State University, PMB 5080, Nkpolu-Oroworukwo, Port Harcourt, Rivers State, Nigeria

Renner Renner
Department of Microbiology, Faculty of Science, Rivers State University, PMB 5080, Nkpolu-Oroworukwo, Port Harcourt, Rivers State, Nigeria

Igoni
Department of Microbiology, Faculty of Science, Rivers State University, PMB 5080, Nkpolu-Oroworukwo, Port Harcourt, Rivers State, Nigeria

Yirabari Gote
Department of Microbiology, Faculty of Science, Rivers State University, PMB 5080, Nkpolu-Oroworukwo, Port Harcourt, Rivers State, Nigeria

Correspondence Author;
Douglas
Department of Microbiology, Faculty of Science, Rivers State University, PMB 5080, Nkpolu-Oroworukwo, Port Harcourt, Rivers State, Nigeria

Isolation of bacteria from soil and water environments with phosphate solubilizing potential

Douglas, Salome Ibietela, Nrior, Renner Renner, Igoni and Yirabari Gote

Abstract
Phosphate-solubilizing bacteria (PSB) are important in the environment due to their role in the Phosphorus cycle, making fixed phosphorus accessible to plants for their use in producing organic acids that dissolve inaccessible phosphate compounds. This study was aimed to isolate and identify bacteria with phosphate solubilizing potential from soil and aquatic environments. Rhizosphere soil samples from around leguminous plant roots and surface water samples from fresh, brackish and marine environments were collected. Using standard Microbiological procedures, these samples were inoculated on Nutrient agar and Pikoskaya's (PVK) medium. Cultural, biochemical and molecular characterization of the isolates were done. The identified isolates were further screened for their ability to solubilize phosphate using the Phosphate Solubilizing Index (PSI). The result of the total heterotrophic bacterial counts (THBC) of the soil sample was 1.2×10^9 CFU/g, freshwater sample was 3.8×10^5 CFU/ml, 6.2×10^6 CFU/ml for the brackish water and 1.4×10^6 CFU/ml for marine sample. Total phosphate solubilizing bacterial counts in the samples were soil (4.8×10^5 CFU/g) > Brackish water (3.1×10^4 CFU/ml) > Marine water (8.8×10^3 CFU/ml) > Freshwater (3.6×10^3 CFU/ml). The populations of these bacteria were observed to be higher in the soil than in the three water samples. The isolates identified as PSB include *Chryseobacterium aquifrigidense*, *Bacillus infantis*, *Pseudomonas xiamenensis*, *Bacillus flexus*, and *Pantoea dispersa*. Results of Zone of clearance on the PVK medium and Solubilizing index (PSI) show that *Pseudomonas xiamenensis* is 3.15±0.06 > *Bacillus flexus* (2.83±0.04) > *Chryseobacterium aquifrigidense* (1.87±0.60) > *Pantoea dispersa* (1.77±0.03) > *Bacillus infantis* (1.43±0.02). It is therefore, recommended that farm soil with a very low soluble phosphate profile be augmented with *Pseudomonas xiamenensis* or *Bacillus flexus* to increase the soluble phosphate content, enhance proper plant growth and increase crop production.

Keywords: Phosphate solubilizing index, rhizosphere, aquatic environment, plant growth

1. Introduction
Phosphorus being an important macronutrient play a crucial role in many metabolic activities including the production of adenosine triphosphate (ATP), phospholipids, formation of nucleic acids, coenzymes, cell division, phosphoproteins, growth of new cells, controls or regulates the synthesis of proteins, growth and development of plants and also involved in energy transfer (Elhaissoufi *et al.*, 2021; Tian *et al.*, 2021; Nrior *et al.*, 2022) [8, 26, 16]. This nutrient is involved in various activities in the cell, including energy production, photosynthesis, utilization of carbohydrates, redox homeostasis, and signaling (Koch *et al.*, 2018) [13]. The presence of this nutrient leads to development of roots, root characteristics, modifications, and root hair density which in turn leads to improved crop production (Elhaissoufi *et al.*, 2021) [8]. Phosphorus is a limiting nutrient for the production of biomass in both soil and water environments. The absence of available Phosphorus in the right quantity in the rhizosphere can produce limitations on normal plant growth, for example leading to the formation of brown leaves and delayed maturity in the plants (Kirui *et al.*, 2022) [11]. Lack of phosphorus in the soil can lead to a decrease (of about 15%) in crop production. Phosphorus fertilizers are the most used fertilizers after Nitrogen (Chakdor *et al.*, 2018). The continuous introduction of inorganic phosphorous fertilizer has led to the eutrophication of the surrounding water bodies. These fertilizers are used in agriculture to improve crop production. However, their availability for crop use may be low because of phosphate fixation to soil cations (Kirui *et al.*, 2022) [11]. They can combine with soil divalent cations including aluminum (Al), iron (Fe), and calcium (Ca) to form fixed soil Phosphate (Shen *et al.*, 2011) [22].

Thus, rhizosphere soils may have large quantities of Phosphate that may not be available for plant use due to failure to be dissolved and liberated in organic form (Elhaissoufi *et al.*, 2021) [8]. Plants prefer water-soluble Phosphates such as monohydrogen phosphate, dihydrogen phosphate and orthophosphate whose concentrations are really small, ranging from 0.001 mg l^{-1} (deficient soil) to 1.0 mg l^{-1} (highly fertile soil); (Kishore *et al.*, 2015) [12].

Research has shown that phosphate-solubilizing microorganisms (PSM) are a large group of organisms that are involved in the bioavailability of phosphorus in the soil (Tian *et al*., 2021) [26]. These groups of organisms play a very important role in the phosphorus cycle, through the decomposition and mineralization of organic phosphate, solubilizing mineral phosphate and storage of huge quantities of P in biomass (Gross *et al*., 2020) [9]. Soil organic phosphorus can increase the phosphate concentration of the soil through the decomposition of biological substances like nucleic acid, phospholipids, phosphoproteins, coenzymes and other organic sources. The presence of PSB can enhance the concentration of inorganic phosphate by mineralization and decomposition of organic phosphate using the phosphatase enzyme (Tian *et al*., 2021) [26]. Bacteria occupy a very prominent position in the Phosphorus cycle enhancing phosphorus availability in the soil ecosystem (Alori *et al*., 2017) [1]. PSB can make this element available for plant use by producing some organic acids and the production of phosphatase enzymes (Elhaissoufi *et al.*, 2021; Tian *et al*., 2021) [8, 26]. Research has shown that PSBs can be applied to soil as an inoculum to elevate the concentration of P present for plant use. PSBs are important because of their ability to make P easily available to plants in both mineral and organic forms through the conversion of fixed Phosphorus compounds, to soluble mineral forms making them more bioavailable (Chen *et al*., 2016) [4]. The release of Phosphate dissolving substances including siderophores, protons, and organic acids, is the requirement for the Phosphate solubilization by organisms such as bacteria, fungi and others. The release of the acids (organic), results in increased availability of phosphorus for plant uptake in the soil through the formation of cation complexes (Al- or Fr-P) or by blocking P absorption sites on soil particles (Elhaissoufi *et al.*, 2021) [8]. Organic acids secreted are due to microbial metabolism, either by oxidative respiration or by glucose fermentation (Demissie *et al.*, 2013) [6].

Some of the bacterial genera that have been identified as PSB include; *Rhodococcus*, *Bacillus*, *Chryseobacterium*, *Pseudomonas*, *Serratia*, *Azotobacter*, *Rhizobium*, *E. coli*, *Phyllobacterium*, *Delftia*, *Xanthomonas*, *Arthrobacter*, *Gordonia* (de Amara *et al.*, 2020; Tian *et al*., 2021) [26]. They have been shown to possess phosphate-solubilizing abilities with plant growth-promoting activities (Taurian *et al.*, 2010; Liu *et al.*, 2015) [25, 14]. This study is aimed at the isolation and molecular identification of bacteria with phosphate solubilizing-potential from soil and aquatic environments.

2. Materials and Methods

2.1 Sample Collection

Composite soil samples were collected from the rhizosphere of leguminous plants. The soil sample was collected from a groundnut farm in Elele town in Etche Local Government Area (LGA), Rivers State, into sterile zip lock bags. The samples were collected from four different parts of the farm, about 5 meters apart, bulked for homogeneity. Water samples were collected from fresh, brackish and marine ecosystems. Fresh water was collected from Bane town in Khana L.G.A; brackish water sample was collected from the new Calabar River, Choba in Obio/Akpor L.G.A while the marine water was collected from Bonny River, in Bonny LGA, all in Rivers State, Nigeria. Surface water samples were collected randomly at four different points at each site and pulled together to form a composite sample. The samples were properly labeled, put in the ice box and immediately transported to the Microbiology Laboratory for analysis (Douglas *et al*., 2018) [7].

2.2 Isolation of the Bacteria

Pikovskaya's agar (PVK) medium containing Glucose, 10g, $Ca_3(PO_4)_2$, 0.5 g, $MgSO_4.7H_2O$, 0.1 g, KCl, 0.2 g, 5 g, $(NH4)_2SO_4$, Yeast extract, 0.5g, $MnSO_4.H_2O$, 0.002 g, $FeSO_4.7H_2O$, 0.001 g, and distilled water, 1 liter was added for the isolation of PSB (Sharon *et al.*, 2016) [21], while Nutrient agar (NA) was used in the isolation of total heterotrophic bacteria. The samples were resuscitated in sterile normal saline (9ml) as the diluents and 10-fold serial dilution was carried out. An aliquot (0.1ml) of the diluted samples were spread on the prepared media plates with the aid of a sterile hockey stick. The inoculated plates were incubated for 24-48 hours at 37 ºC. After culture incubation, the total heterotrophic bacterial counts were determined by counting the colonial growth on the plates, and the CFU/g (colony forming unit per gram) was calculated. Various colonies that developed on the plates were sub-cultured on NA and PVK plates. This was achieved by streaking distinct colonies based on their cultural characteristics until pure isolates is obtained. The pure colonies were stored in agar slants and refrigerated at 4 ºC, until required for use (Nrior *et al*., 2022) [16].

2.3 Biochemical and Molecular Test for Identification

The following biochemical test was carried out; Motility, Methyl Red, Voges-Proskauers (MRVP) test, sugar fermentation tests, catalase, oxidase, indole, production test, test for hydrogen sulfide and gas production, citrate utilization test, and urease test. Microscopy of the isolates using the Gram Staining technique was also carried out (Cheesbrough, 2005) [3].

Molecular characterization of the isolates was carried out by extracting the DNA fragment, amplification and sequencing the 16S rRNA gene (Ogbonna and Azuonwu, 2019) [17].

2.4 Screening of the Bacterial Isolates for the Phosphate Solubilizing Potential

Adopting the method of Sharon *et al.*, (2016) [21] the isolates from the initial PVK media plates were inoculated on a freshly prepared PVK agar medium with the use of an inoculating needle and incubated for 4 to 7 days for better analysis of the zone of clearance. The bacteria with phosphate solubilizing potential produced a clear zone of inhibition which was measured and the phosphate solubilizing index (PSI) was then calculated using the formula by Pande *et al*., (2017) [18].

$$PSI = \frac{\text{colony diameter} + \text{clearance zone}}{\text{Colony diameter}} \quad \ldots\ldots\ldots\ldots\ldots equation\ 1$$

3. Results

3.1 Bacterial Population

The results of the bacterial counts in the soil and three water habitats are presented in Table 1. The THBC of the soil sample was 1.2×10^9 CFU/g, the THBC of the freshwater sample was, 3.8×10^5 CFU/ml, 6.2×10^6 CFU/ml for the brackish water sample and 1.4×10^6 CFU/ml was recorded in the marine sample with the Freshwater samples having the least heterotrophic bacterial counts while the soil sample had the highest counts of total heterotrophic bacterial counts. The counts of culturable phosphate solubilizing bacteria (on PVK medium) was recorded as 4.8×10^5 CFU/g for the soil sample, 3.6×10^3 CFU/ml for fresh water sample, 3.1×10^4 CFU/ml for the brackish water and 8.8×10^3 CFU/ml for the marine water sample as the fresh water recorded the lowest count of phosphate-solubilizing bacteria while the highest count was observed in the soil sample.

3.2 Biochemical Characteristics of the Bacterial Isolates

The isolates were characterized based on the biochemical test results presented in Table 2. The isolates were identified by comparing them with the standards provided in Bergey's Manual of Determinative Bacteriology (Holt *et al*., 1994) [10], for identification. They were tentatively identified as; *Chryseobacterium* sp., *Bacillus* sp., *Pantoea* sp. and *Pseudomonas* spp,

Table 1: Total Heterotrophic and Phosphate Solubilizing Bacterial Counts in the Samples

Samples	Bacterial Population	
	Total Heterotrophic Bacteria (THB)	**Phosphate solubilizing Bacteria (PSB)**
Soil	1.2×10^9 CFU/g	4.8×10^5 CFU/g
Fresh water	3.8×10^5CFU/ml	3.6×10^3 CFU/ml
Brackish water	6.2×10^6 CFU/ml	3.1×10^4 CFU/ml
Marine water	1.4×10^6 CFU/ml	8.8×10^3 CFU/ml

Table 2: Microscopic and Biochemical Characteristics of the Isolates Obtained From the Samples

Isolates code	Grams reaction	Cell morphology	Catalase	oxidase	Indole	MR	Urease	Citrate	Motility	TSIA				Starch hydrolysis	Glucose	Sucrose	Lactose	Probable organisms
										Butt	slant	Gas	H_2S					
PSB 1	-	Rod	+	-	-	-	-	+	-	B	B	-	-	-	-	-	-	*Pseudomonas* sp
PSB 2	+	Rod	+	-	-	-	-	+	-	B	B	-	-	-	-	-	-	*Bacillus* sp
BE2	+	Rod	+	+	-	-	-	+	-	B	B	-	-	-	-	-	-	*Bacillus* sp
PSB 17	-	Rod	+	+	-	+	+	+	+	B	A	-	-	-	+	+	+	*Pseudomonas* sp
PSB 20	+	Rod	+	+	-	-	-	-	+	B	A	-	-	+	-	-	-	*Bacillus* sp
BB5	-	Rod	-	-	-	-	-	+	-	B	B	-	-	-	-	-	-	*Pantoea* sp
SE2	-	Rod		+	+	-	+	-	-	B	B	-	-	-	-	-	-	*Chryseobacterium* sp

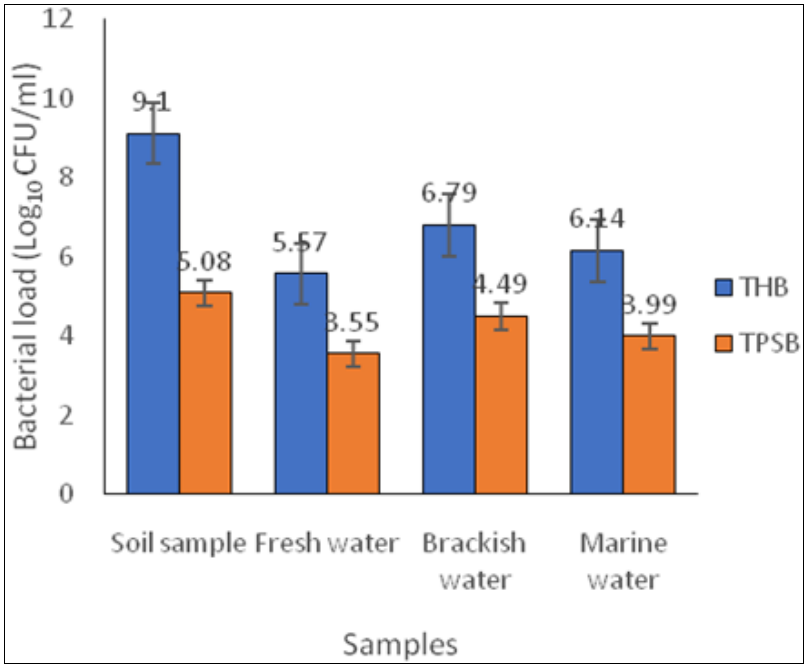

Fig 1: Total Heterotrophic and Phosphate Solubilizing Bacterial counts of the samples

Table 3: Screening for Phosphate Solubilizing

Isolates	Zone of clearance on the PVK medium		Solubilizing index (SI) (A/B)
	A(mm)	B(mm)	
Bacillus flexus	8.95±0.07	3.15±0.07	2.83±0.04
Pseudomonas xiamenensis	8.05±0.00	2.55±0.07	3.15±0.06
Pantoea dispersa.	7.1±0.14	4±0.00	1.77±0.03
Bacillus infantis	8.5±0.7	5.5±0.00	1.43±0.02
Chryseobacterium aquifrigidense	5±0.00	2.05±0.07	1.87±0.60

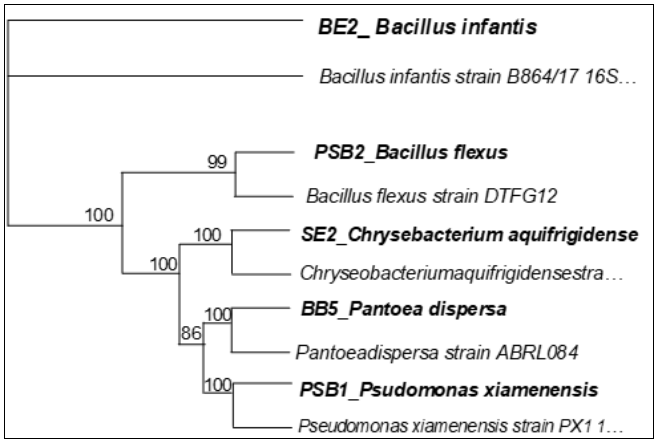

Fig 2: The Phylogenetic tree of the identified Isolates organisms

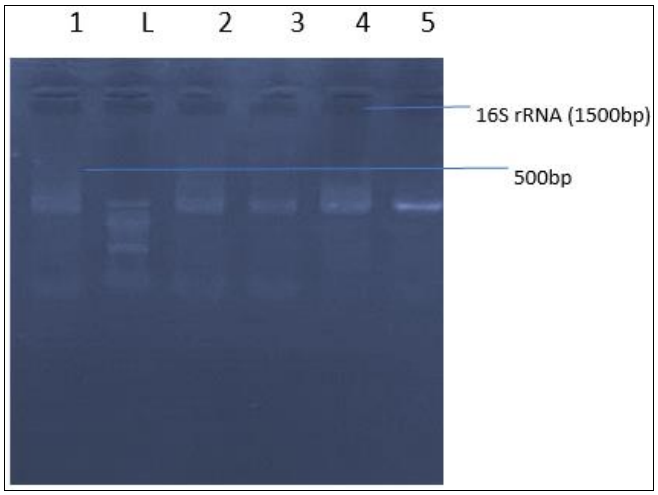

Fig 3: Agarose gel electrophoresis of the 16S rRNA of the bacterial isolates.

3.3 Molecular Results

The phylogenetic tree, in Figure 2 shows the relationship of the isolates to different species of the genus. The obtained 16S rRNA sequence from the isolates produced an exact match during the mega blast search for highly similar sequences from the NCBI non-redundant nucleotide (nr/nt) database. The 16S rRNA of the isolate PSB1 showed a percentage similarity to other species at 100% relatedness. The evolutionary distances computed using the Jukes-Cantor (1969) method agreed with the phylogenetic placement of the 16S rRNA of the isolates within the *Pseudomonas* sp and revealed a close relatedness to *Pseudomonas xiamenensis* strain PX1. The 16S rRNA sequence of the isolate BE2 showed 100% relatedness to *Bacillus infantis* strain B864/17, and the 16S rRNA of the isolate PSB 2 showed 99% relatedness to *Bacillus flexus* strain DTFG12. The sequence of 16S rRNA of the isolate, SE2 recorded a 100% relatedness to the organism, *Chryseobacterium aquifrigidense* strain CW9 while the 16S rRNA of the isolate BB5 showed a 100% relatedness to the bacterium, *Pantoea dispersa* strain ABRL084. The results of the agarose gel electrophoresis of the 16S rRNA gene as seen in Figure 3 shows the base pair of the bacterial isolates analyzed. Lanes 1-5 show the amplified 16S rRNA bands while lane L represents the 100 bp ladder.

3.4 Phosphate Solubilizing Index

The isolates that showed the potential to solubilize phosphorus or phosphate produced a halo zone of clearance on the PVK medium as shown in Figure 4. The measure of the diameter of the halo zone to the growth of the bacteria was used in the determination of the phosphate solubilizing index. The results of the phosphate solubilizing index (PSI) carried out on the isolates are shown in Table 3. Analysis of the Zone of clearance on the PVK medium and SI shows that *Pseudomonas xiamenensis* is (3.15±0.06) > *Bacillus flexus* (2.83±0.04) > *Chryseobacterium aquifrigidense* (1.87±0.60) > *Pantoea dispersa* (1.77±0.03) > *Bacillus infantis* (1.43±0.02).

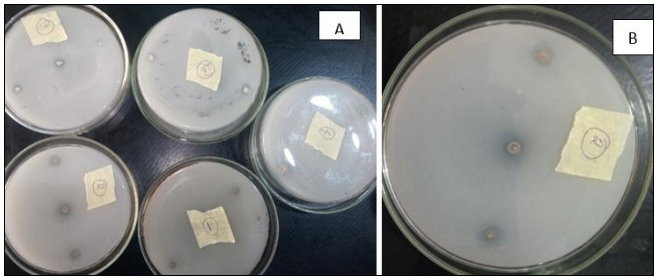

Fig 4: The screened plates of the different isolates of bacteria with phosphate solubilizing potential

4. Discussion

The Total Heterotrophic bacterial counts (THBC) were highest (1.27×10^9 CFU/g) in the soil sample compared to the aquatic ecosystems analyzed. The high THBC of the soil sample was followed by the brackish water with counts of 6.2×10^5 CFU/ml followed by the marine water which recorded counts of 1.4×10^6 CFU/ml and the least was recorded as 3.8×10^5 CFU/ml in the freshwater sample. The higher total THBC in the soil sample may be attributed to the availability of nutrients, the favourable and stable conditions, favouring bacterial growth in the soil in comparison to the other samples. In addition, most of the bacteria present in the aquatic habitat come from the terrestrial habitat through run-off. This observation is similar to the reports of Mahalakshmi *et al*., (2011) [15] who reported highest total heterotrophic bacterial counts in the sediments than the water samples. Phosphate solubilizing bacterial counts in the samples, as shown in Figure 1, were higher in the soil, too: ($4.8\text{x}10^5$ CFU/g) > Brackish water ($3.1\text{x}10^4$ CFU/ml) > Marine water ($8.8\text{x}10^3$ CFU/ml) > Freshwater ($3.6\text{x}10^3$ CFU/ml). High populations of phosphate solubilizing bacterial counts in the soil can be attributed to the role they play in the soil to ensure the availability of phosphorus through phosphate cycling and other nutrients for the enhancement of soil fertility (Elhaissoufi *et al*., 2021; Kishore *et al*., 2015) [8, 12]. The high counts of PSB in the soil is consistent with the study of Asuming-Brempong and Aferi, (2014) [2] in which high counts of phosphate solubilizing bacteria were also recorded in soil samples.

The isolated, identified, and screened PSB in this study include: *Chryseobacterium aquifrigidense* strain CW9, *Bacillus flexus* strain DTFG12, *Pantoea dispersa* strain ABRL084 *Bacillus infantis* strain B864/17 and *Pseudomonas xiamenensis* strain PX1. These were similar to those obtained in other studies (Assuming-Brempong and Aferi, 2014; Sharon *et al*., 2016; Singh *et al*., 2013) [2, 21, 23]. Chakdor *et al*., (2018), identified the following as phosphate solubilizing bacteria: *Bacillus* sp. AH9, *Kosakonia* sp. A37, *Pantoea* sp. A34, *Pantoea* sp. A3, *Kosakonia* sp. B7, as

highly efficient from a termite mound soil. *Kosakonia* sp. identified by them was not identified in this study.

The phosphate-solubilizing index (PSI) of the identified bacteria as shown in Table 3 ranged from 1.34±0.01 to 3.15±0.06 which was similar to the index range of the phosphate-solubilizing bacteria screened by Sitepu *et al.*, (2014) [24] in which the PSI values of similar bacteria were within 1.2 to 3.7 in a study to isolate potential PSB from peat swamp forest Central Kalimantan. However, the PSI values observed in the present study were lower than what was observed by Pande *et al.* (2017) [18] to characterize PSB and their efficacy on the growth of maize in which the PSI values were above 4.0. The isolate with the highest PSI value was *Pseudomonas xiamenensis* followed by *Bacillus flexus* which is closely related to the observations of the studies of Jha *et al*., (2009) and Sharma *et al.,* (2013) [20] which identified the genera, *Pseudomonas* and *Bacillus* among other bacteria with higher phosphate solubilizing potential in a study to determine the phosphate solubilization ability and antimicrobial potential of a novel florescent *Pseudomonad* strains. The study of Alori *et al.*, (2017) [1] also identified the genera, *Bacillus* and *Pseudomonas* as phosphate solubilizers. Chakdor *et al*., (2018), showed *Bacillus* sp. to give the highest PSI of 3.5 on PKV agar. *Pseudomonas* and *Bacillus* spp. are among the frequently encountered PSBs identified. *Bacillus* spp. due to its capacity to form spores and has been used to produce biofertilizers since it can tolerate harsh environmental conditions and remain in the soil environment for a prolonged period. In another study by Sarker *et al*., (2014) to isolate phosphate-solubilizing bacteria that enhance growth and improve nutrient uptake by wheat, the genus, *Pseudomonas* was also isolated and identified to have better phosphate solubilizing potential, hence increasing the growth of wheat. The greater potential of *Pseudomonas* sp to solubilize phosphate may be due to its capacity to produce more organic acid during microbial metabolism (Demissie *et al.*, 2013) [6]. The lower PSI value is an indication of the lower ability to solubilize Phosphate.

According to Assuming-Brempong and Aferi, (2014) [2], different bacteria use different pathways to produce different organic acids with a chelating ability, hence their solubilizing potential. *Chryseobacterium* sp according to previous studies is a good rhizosphere colonizer because of its affinity with the root thus enhancing plant growth (Singh *et al.*, 2013) [23]. The findings of Zaidi and Khan, (2006) identified *Chryseobacterium* sp. as a phosphate-solubilizing bacteria and growth enhancer. *Pantoea dispersa* was also identified as an efficient PSB isolated from termitarium soil (Chakdor *et al*., 2018). *Pantoea* sp is a diverse group of pigmented (yellow) bacteria with growth enhancement ability for plants and have been frequently isolated from both terrestrial and aquatic environments. They exhibit antimicrobial activity, phosphate-solubilizing, and bioremediation potentials (Walterson and Stavrinides, 2015) [27]. According to another study by Sharon *et al*., (2016) [21], *Pantoea* sp was identified as a phosphate-solubilizing bacteria and growth enhancer of plants.

5. Conclusion

This research has revealed the presence of phosphate-solubilizing bacteria in soil and water ecosystems and has shown that their population in the soil samples was higher, compared to the aquatic ecosystems. The following bacteria were identified as PSB after screening: *Bacillus flexus*, *Bacillus infantis*, *Pseudomonas xiamenensis*, *Pantoea dispersa* and *Chryseobacterium aquifrigidense*. Results obtained from this study, has further revealed that *Pseudomonas xiamenensis* is a better phosphate-solubilizing bacterium, since it had a higher PSI.

It is recommended that these species isolated from this study could be further evaluated to ascertain their effectiveness and utilization as biofertilizers in the field to increase soil fertility and crop productivity. However, farm soil with a very low soluble phosphate profile may be augmented with *Pseudomonas xiamenensis* or *Bacillus flexus* to enhance proper plant growth and increase crop production.

6. Conflict of Interest

Not available

7. Financial Support

Not available

8. References

1. Alori ET, Glick BR, Babalola OO. Microbial phosphorus solubilization and its potential for use in sustainable agriculture. Front Microbiol. 2017;8:971-982.
2. Assumed-Brempong S, Aferi NK. Isolation of phosphate solubilizing bacteria from tropical soil. Glob Adv Res J Agric Sci. 2014;3(1):8-15.
3. Cheesbrough M. District laboratory practice in tropical countries, part 2. Cambridge: Cambridge University Press; c2005. p. 159-162.
4. Chen W, Yang F, Zhang L, Wang J. Organic acid secretion and phosphate solubilizing efficiency of Pseudomonas sp. PSB12: Effects of phosphorus forms and carbon sources. Geomicrobiol J. 2016;33:870-877.
5. De Amaral LA, de Souza CAA, de Almeida LR, de Oliveira-Longatti SM, Lustosa FJF, de Souza MF. Selected bacterial strains enhance phosphorus availability from biochar-based rock phosphate fertilizer. Ann Microbiol. 2020;70:1-13.
6. Demissie S, Muleta D, Berecha G. Effect of phosphate solubilizing bacteria on seed germination and seedling growth of Faba bean (*Vicia faba* L.). Int J Agric Res. 2013;8:123-136.
7. Douglas SI, Nrior RR, Kporman LB. Toxicology of spent phone bacteria on microflora in marine, brackish, and freshwater ecosystems. J Adv Microbiol. 2018;12(2):1-10.
8. Elhaissoufi W, Gheulam C, Barakat A, Zeroul Y, Bargaz A. Phosphate bacterial solubilization: A key rhizosphere driving force enabling higher P use efficiency and crop productivity. J Adv Res. 2021;8(14):090-123.
9. Gross A, Lin Y, Weber PK, Pett-Ridge J, Silver WL. The role of soil redox conditions in microbial phosphorus cycling in humid tropical forests. Ecology; c2020 .p. 101.
10. Holt JG, Krieg NR, Sneath PH, Staley JT, Williams ST. Bergey's manual of determinative bacteriology. Baltimore: Williams and Wilkins; c1994. p. 151-157.
11. Kirui CK, Njeru EM, Runo S. Diversity and phosphate solubilization efficiency of phosphate solubilizing bacteria isolated from semi-arid agroecosystem of Eastern Kenya. Microbiol Insights. 2022;15:1-12.

12. Kishore N, Pindi PK, Reddy SR. Phosphate-solubilizing microorganisms: A critical review. Telangana, India; c2015. p. 307-313.
13. Koch M, Kruse J, Eichler-Löbermann B. Phosphorus stocks and speciation in soil profiles of a long-term fertilizer experiment: Evidence from sequential fractionation, P K-edge XANES, and 31P NMR spectroscopy. Geoderma. 2018;316:115-126.
14. Liu Z, Li YC, Zhang S, Fu Y, Fan X, Patel JS. Characterization of phosphate solubilizing bacteria isolated from calcareous soils. Appl Soil Ecol. 2015;96:217-224.
15. Mahalakshmi M, Srinivasan M, Murugan M, Balakshnan S, Devanathan K. Isolation and identification of total heterotrophic bacteria and human pathogens in water and sediment from Cuddalore fishing harbour after the Tsunami. Asian J Biol Sci. 2011;4(2):148-156.
16. Nrior RR, Douglas SI, Igoni YG. Toxicity of herbicide, paraquat dichloride and insecticide, lambda-cyhalothrin on phosphate solubilizing bacteria, Pantoea dispersa in aquatic ecosystems. Microbiol Res J Int. 2022;32(4):13-21.
17. Ogbonna DN, Azuonwu TC. Plasmid profile and antibiotic resistance pattern of bacteria from abattoirs in Port Harcourt City, Nigeria. Int J Pathogen Res. 2019;22:1-11.
18. Pande A, Pandey P, Mehra S, Singh M, Kaushik S. Phenotype and genotypic characterization of phosphate solubilizing bacteria and their efficacy on the growth of maize. J Genet Eng Biotechnol. 2017;15:379-391.
19. Saker A, Talukder NM, Islam T. Phosphate solubilizing bacteria promote growth and enhance nutrient uptake by wheat. Plant Sci Today. 2004;1(2):86-89.
20. Sharma SB, Sayyed RZ, Trivedi MH, Gobi TA. Phosphate solubilizing microbes: Sustainable approach for managing phosphorus deficiency in agricultural soils. Springerplus. 2013;2:587.
21. Sharon JA, Glenn GM, Imam SH, Lee CC. Isolation of efficient phosphate solubilizing bacteria capable of enhancing tomato growth. J Soil Sci Plant Nutr. 2016;16(2):525-536.
22. Shen J, Yuan L, Zhang J, Li H, Bai Z, Chen X, Zhang W, Zhang F. Phosphorus dynamics: from soil to plant. Plant Physiol. 2011;156:997-1005.
23. Singh AV, Chandra R, Goel R. Phosphate solubilization by *Chryseobacterium* and their combined effect with N and P fertilizer on plant growth promotion. Arch Agron Soil Sci. 2013;59(5):641-651.
24. Sitepu RI, Hashidoko Y, Santoso E, Tahara S. Potent phosphate solubilizing bacteria isolated from Dipterocarps grown in peat swamp forest in Central Kalimantan and their possible utilization for biorehabilitation of degraded peatland. Indones J Forestry Res. 2009;6(2):96-118.
25. Taurian T, Anzuay MS, Angelini JG, Tonelli ML, Ludueña L, Pena D. Phosphate-solubilizing peanuts associated bacteria: screening for plant growth-promoting activities. Plant Soil. 2010;329:421-431.
26. Tian J, Ge F, Zhang D, Deng S, Liu X. Roles of phosphate solubilizing microorganisms from managing soil phosphorus deficiency to mediating biogeochemical P cycle. Biology. 2021;10:158.
27. Walterson MA, Stavrinides J. Pantoea: Insight into a highly versatile and diverse genus within the Enterobacteriaceae. FEMS Microbiol Rev. 2015;39(6):968-984.

How to Cite This Article
Douglas, Ibietela S, Nrior, Renner R, Igoni, Gote Y. Isolation of bacteria from soil and water environments with phosphate solubilizing potential. Journal of Advances in Microbiology Research. 2024; 5(2): 145-150.