

Phosphate Solubilising Activity of Soil Microbes Isolated from the Rhizosphere of Tasar growing Field

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Research Article

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ABSTRACT

Phosphorus is an essential element after nitrogen needed for development and growth of plant. P deficiency has adverse effect on the plant such as browning of leaves, leaf size decreases, stem become weak and development become slowly. Phosphate Solubilizing Bacteria present in the soil converts phosphate into bioavailable form through mineralization and solubilisation process. Hence, the study was carried out to study the Phosphate Solubilizing Efficiency by drop plate method of 21 strains isolated from Tasar growing region and to identify the strain through biochemical characterization. It was found that C13 showed the highest PSE of 2.66 followed by C18, 2.14; C11, 1.85; C20,1.71 and C21,1.625. Some of the colonies do not shown any Phosphate Solubilizing Activity C1, C8, C9 and C19. From the result of biochemical study, the strains were identified as *Pseudomonas* spp, *Bacillus* spp, *Planococcus* spp and *Azotobacter* spp.

INTRODUCTION

Silk the "Queen of Textiles" spearheading a silent revolution by providing livelihood to millions. By transferring money from rich to poor and urban to rural. Which is benefitting the poorest of the poor. Thus, it's important to sustain its growth and continue to available for mankind at any cost. India is the only country which produces all the five known commercial silks namely Mulberry, Tropical Tasar, OakTasar, Eri and Muga. Continuous use of inorganic fertilizer has deteriorated the soil health thus affecting the production of cocoon. N, P and K are the major rate limiting nutrient affecting the development of plants. After nitrogen, phosphorus is the major plant growth-limiting nutrient which is abundant in soil in both organic and inorganic. Deficiencies are circumventing by adding chemical fertilizer. 75-90% of fertilizer are precipitated by Fe, Al and Ca complexes cannot be utilized by plant. Phosphorus is an

essential macronutrient needed for growth and development of plants by inducing the physiological activities such as cell division, macromolecular biosynthesis, photosynthesis, respiration of plants, and development of good root system and utilization of carbohydrates. Deficiency of phosphorus lead to browning of leaves accompanied by decrease in the leaf size, weak stem and slow development. Phosphate solubilizing bacteria present in the rhizosphere solubilises phosphorus from insoluble forms by secreting organic acid making them to available forms such as HPO_4^{2-} and H_2PO_4 to plant roots for growth and development. Although chemical fertilizer increases the production by fifty percent but also causes environmental pollution and health hazards ^[1].

The study was carried out to determine the ability of 23 strains to mobilize inorganic and organic P. The 23 strains were cultured in medium to estimate the phosphate solubilizer population by clear zone around the colonies indicating phosphate solubilisation. Enzyme Phosphatase is secreted by Phosphate Solubilising Bacteria which cleave phosphate.

MATERIALS AND METHODS

Soil sample were collected from the 05cm to 17cm depth from the rhizospheric zone of different plant of Tasar growing field of Central Tasar Research and Training Institute, Nagri, Ranchi. All the samples were collected in polythene bags and transported to laboratory aseptically and maintained and stored at 4°C for further analysis. The sample were air dried and grounded to pass through 2 mm sieve before analysis.

Isolation of bacteria and biochemical characterization

Strain of bacteria were isolated from soil sample of Tasar growing field and 02 strain were isolated from biofertilizer bought from BAU, Ranchi. 1gm of soil sample were serially diluted and spread plate in triplicates on NAM media containing Beef extract 3.0g; Peptone 5.0g; Agar 15.0g and Distilled water 1000ml; pH 6.8±0.2. For inoculation 1gm soil sample were diluted with 9 ml of sterilized distilled water and shaken for 15 minutes ^[2].

For inoculation 0.1ml soil dilution sample was taken as inoculums on the media and was spreaded on the media. The plate were incubated at 30°C for 24-48 hrs. Bacterial colonies were isolated based on the morphology of colony in respective medium by agar slant preparation. Repeated sub-cultures were made for pure culture and the colonies are designated as C1, C2, C3, C4, C5.....C23.

Biochemical characterization

Some of the Biochemical Character were studied to identify the bacteria such as Microscopic study, Glucose, Lactose, Sucrose and Mannitol Fermentation, Triple Sugar Test, Starch Hydrolysis, Catalase Activity, Methyl Red Test, Citrate Test, VP test, Litmus Milk Test, Indole test, Urease test, Gelatine Hydrolysis test, Nitrate Reduction Test etc. were determined and characterization were done as per Bergey's manual of determinative bacteriology ^[3].

Phosphate solubilizing activity on solid media

For determination of phosphate solubilising activity of isolated bacteria drop plate method was used. Pikovskaya (PVK) medium containing glucose,10g;Ca₃(PO₄)₂, 5g;(NH₄)₂. SO₄,0.5g; NaCl,0.2g; MgSO₄7H₂O,0.1g; KCl,0.2g; Yeast extract, 0.5g,MnSO₄H₂O,0.002g and FeSO₄7H₂O,0.002g was poured in the Petri plates. The Petri plates were allowed to solidify then the plates were divided into four parts, in each part a loop full of bacterial culture were inoculated in the four part of the plate and same was repeated further for the other isolate. The plate were inoculated at 30°C for 7 days. Colonies of PSB were detected by clear zone of solubilisation [4].

Phosphate solubilisation efficiency

Phosphate Solubilisation Efficiency (PSE) was identified by measuring the total halazone of the colony and the colony diameter.

$$\text{PSE} = \frac{\text{Colony diameter} + \text{halozone diameter}}{\text{Colony diameter}}$$

RESULT AND DISCUSSION

Totally 21 isolates were obtained from soil sample collected from Tasar growing region and the 21 samples were screened for phosphate solubilising activity. Phosphate solubilisation Index (PSI) ranged from 1.2 to 2.66 on PVK media. Microbial colonies showing clear halozones around the microbial growth were considered as phosphate solubilisation by the bacterial colonies.

Colonies isolated from the bio-fertilizer showed the highest PSE i.e. 3.57 and 3.33. From the isolate it was found that colony no 13 showed the highest clear halozones followed by colony no C18, C11, C20, and C21. Some of the colonies does not shown any phosphate solubilising activity like C1,C8,C9 andC19. The phosphate solubilisation index of the isolate are shown in Table 1.

The bacterial isolates were chacterized by series of biochemical reaction and identified as Pseudomonas sp, Bacillus sp,Planococcus sp, Azotobacter spetc. many researchers have identified the genus Azotobacter sp,Bacillus sp and Pseudomonas sp as phosphate solubiliser and also improve the plant growth as reported.

The morphological and biochemical characteristic of the isolates It was concluded from the present study that phosphate solubilising bacteria have important advantage toward soil fertility, sustainability and production of good quality cocoon. However, researches are still limited for screening PSM performant strains further investigation are needed (Figure 1) [5].

Figure 1. A,B,C,D Showing phosphate solubilisation efficiency.

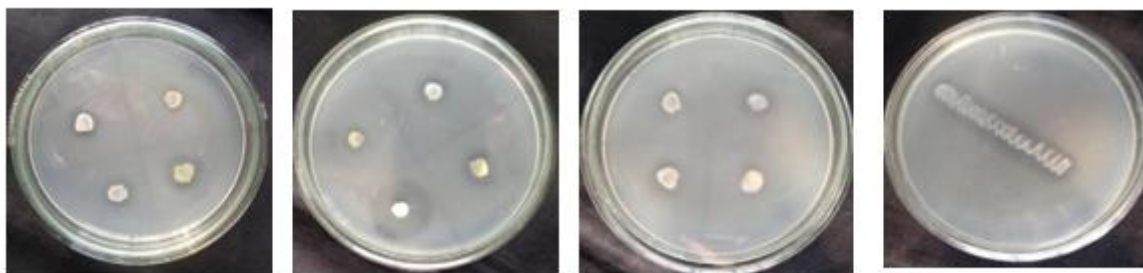


Table 1. Showing morphology and phosphate solubilising efficiency of isolates.

Strain Nos.	Gram staining	Shape	Phosphate Solubilising Efficiency
C ₁	+	Rod	-
C ₂	-	Rod	1.75
C ₃	+	Rod	1.2
C ₄	+	Rod	1.5
*C ₅	-	Rod	3.57
C ₆	+	Rod	1.2
C ₇	-	Rod	1.33
C ₈	+	Cocci	-
C ₉	+	Rod	-
C ₁₀	+	Rod	1.57
C ₁₁	+	Rod	1.85
C ₁₂	+	Rod	1.6
C ₁₃	+	Rod	2.66
C ₁₄	+	Rod	1.5
C ₁₅	+	Rod	1.66
C ₁₆	+	Rod	1.2
*C ₁₇	+	Rod	3.33
C ₁₈	+	Rod	2.14
C ₁₉	-	Rod	-
C ₂₀	+	Rod	1.71
C ₂₁	+	Rod	1.625
C ₂₂	+	Rod	1.5
C ₂₃	+	Rod	1.4

CONCLUSION

It was concluded from the present study that phosphate solubilising bacteria have important advantage toward soil fertility, sustainability and production of good quality cocoon. However, researches are still limited for screening PSM performant strains further investigation are needed.

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REFERENCES

1. Ahemed M, et al. Effect of fungicides on plant growth promoting activities of phosphate solubilising pseudomonas putida isolated from mustard (brassica campestris) rhizosphere. Chenaosphere. 2012;86;945-950.
2. Zaidi A, et al. Biological importance of phosphorus and phosphate solubilising microorganisms: An overview. 2009.
3. Kothamasi D, et al. Arbuscular mycorrhizae and phosphate solubilising bacteria of the rhizosphere of the mangrove ecosystem of Great Nicobar island, India, Biol Fertil Soils.2006;42:358-361.
4. Edi-premono, et al. Effect of Phosphate Solubilizing Pseudomonas putida on the growth of Maize and its survival in the Rhizosphere. Indo J Crop Sci. 1996;11:13-23.
5. Alikhani HA, et al. Phosphate Solubilization activity of Rhizobia native to Iranian Soils. Plant Soil. 2006;287:35-41.