Available online at www.pelagiaresearchlibrary.com

Pelagia Research Library

European Journal of Experimental Biology, 2013, 3(2):191-193

ISSN: 2248 –9215
CODEN (USA): EJEBAU

Study of influence of phosphate dissolving micro-organisms on yield and phosphate uptake by crops

Shirish S. Pingale[1] and Popat S. Virkar[2]

[1]*P.G. Department of Chemistry, Gramonnati Mandal's Arts, Commerce and Science College, Narayangaon, Pune, Maharashtra, INDIA*
[2]*P.G. Department of Chemistry, C. T. Bora College Shirur, Dist-Pune (Affiliated to University of Pune)*

ABSTRACT

The use of phosphate solubilizing bacteria as inoculants simultaneously increases Phosphorus uptake by the plant and crop yield. Strains from the genera Pseudomonas, Bacillus and Rhizobium are among the most powerful phosphate solubilizers. The principal mechanism for mineral phosphate solubilization is the production of organic acids, and acid phosphatases play a vital role in the mineralization of organic phosphorous in soil. Several phosphatase-encoding genes have been cloned and characterized and few genes involved in mineral phosphate solubilization have been isolated. Therefore, genetic manipulation of phosphate-solubilizing bacteria to improve their ability to enhance plant growth may include cloning genes involved in both mineral and organic phosphate solubilization, followed by their expression in selected rhizobacterial strains. The extent to which soil phosphorus (P) status gets affected the incidence of soil phosphate-solubilising bacteria (PSB). Bacteria were isolated from rhizosphere and non-rhizosphere soils differing in Phosphorus status. The P-solubilising phenotype was determined on agar supplemented with sparingly-soluble mineral phosphates ($Ca_2OH(PO_4)_3$ and $CaHPO_4$). The frequency of P-solubilisation in the bacterial population was significantly greater in soils of low-P status. P-solubilising bacteria from high-P level soils and soils which had not received P fertiliser were identified. The phytogenetic composition of PSB differed significantly ($P < 0.05$) between sites, however nearly half the families were common across all sites, constituting a 'core community' of P-solubilising bacteria. As the abundance and composition of P-solubilising bacteria are under strong selection pressure affected through farm management strategies, better understanding of their ecology provides the opportunity to increase the availability of soil P for plant-uptake.

Keywords: phosphate solubilizing bacteria, mineral phosphate solubilization, Rhizobium, rhizobacterial strains.

INTRODUCTION

Determination of the effect of bio-fertilizer on crop of maize and potato, especially phosphate solubilizing micro-organisms. Though most soils contain appreciable amounts of inorganic phosphate, most of it being in insoluble forms, cannot be utilized by crops unless they are solubilized. Soils also contain organic P that could be utilized by plants only when it is mineralized. Phosphate solubilizing organisms are not able to solubilize insoluble forms of inorganic P but are also capable to mineralize organic forms of Phosphorus thus improving the availability of native soils Phosphorus making their available to plants. Phosphate solubilizing microorganism bioferitilizer being

economical and environmentally safe offers a viable alternative to the chemical fertilizers. Both inorganic and organic forms of phosphorus are important to plants as sources of Phosphorus. The relative amounts in the two forms vary greatly from soil to soil, but it is not at all uncommon for more than half the phosphorus to be in the organic form. Although organic P in soil is somewhat biologically stable, continuous turnover occurs through mineralization Immobilization and the equilibrium level will depend to a considerable extent on the nature of the soil and its environment[1, 2]. The Phosphorus content of soil organize matter ranges from <3.0%, which is reflected in variable C/P ratio of soils. A lower c/p ratio has been reported to associate with the lower molecular weight componenets of soil organic matter. The experimental method with field work was carried out successfully within the stipulated time period. The important outcome of this work is summarized as follows[3, 4].

MATERIALS AND METHODS

Isolation of phosphate sulubilizing micro-organism a known quality of soil rhizosphere is suspended in a known volume of sterile water and serial dilutions of the suspension made in sterile blanks. Appropriate dilutions are plated on phosphate containing solid media for obtaining microorganisms capable of dissolving phosphate. The plates are incubated for 4-5 days. Such cultures were isolated, identified and the extent of solubilisation will be determined quantitatively. Katzne/ Smand Bose medium, Pikoveskay's medium etc[5].

A. **Quantitative measurement of phosphate solubilization in culture medium**: Selected culture are grown in 50-100 ml. Aliquots of Pickovskyaya's liquid medium for 6-17 days at 28^0C. In the case of fungi the Whatmann No. 42 filter paper. Due to pigments, the filtrate may often be colored with 1-2 gm. of cultivated charcoal to remove the color impurities. Whatmann No. 1 paper is used for further filtration and passing through centrifuse machine at 10,000/- r.p.m. for 10-15minute. In 10 ml. Aliquot 2-5 ml. of barton's reagent is added and after 10 minute the intensity of color is reported in a colorimeter using 430nm wavelength and calculated the strength of solution.

B. Solubilization of phosphates by Microorganisms: Belonging to the genera pseudomonas and bacillus and fungi belonging to the genera penicillium and aspergillus posses the ability to bring insoluble phosphates in soil into soluble forms by secreting organic in soil into soluble forms by secreting organic acids such as formic acids, aceticacid, fumaric acid, succinic acids etc. The acids lower the pH and bring about the dissolution of bound forms of phosphate. Some of the hydroxyl acids may chelate with calcium and iron resulting in effective solabilization and utilization of phosphates. Solubilization of $Ca_3 (PO_4)_2$ by fungi associated with root nodules

Genera	P_2O_5, mg	% solubilized	pH of filrate
A)Penicillium lilacinum	+8.87	39.02	4.7
B) Aspergillus sp	+4.04	17.77	5.6
C) Aspergillus niger	+2.07	9.10	8.6
D) Rhitoctonia solani	-1.77	0.00	8.1

C. Reagents used

a) Pikovskyaya's medium

Glucose	10.0g
$Ca_3(PO4)2$	5.0g
$(NH_4)_2 SO_4$	0.5g
KCl	0.2g
$MgSO_4. 7 H_2O$	0.5g
$MgSO_4$	trace
$FeSO_4$	trace
Yeast extract	0.5g
Agar	15.0g
Distilled water	1000ml

b) Barton's reagent

A = 25gm ammonium molybdate to be dissolved in 400ml. H_2O.

B = 1.25 gm ammouium metavanadate in 300ml of boiling water, cooled and then 250 ml. Conc. HNO_3 added. Afterwards, A and B solutions are mixed and made up to a liter.

D .Quantitative Measurement of Phosphate solubilization in culture medium.
Selected cultures are grown in 50-100 ml aliquots of pikovskyaya's liquid medium for 6-17 days at 30^0C. In the case of fungi, the culture is filtered using whatmann filter paper No. 42.Then the culture is treated with activated charcoal to eliminate the pigment colored impurities. Then the bacterial cultures are filtered through whatmann filter paper No. 1 to remove the insoluble phosphate and centrifuged at 1000 rpm. for few minutes. Filtration and centrifugation is repeated until clear solution is obtained. To a 10 ml aliquot of the neat filtrate 2.5 ml of Barton's reagent is added and the volume made up to 50 ml. After 10 minute the resultant color is read in a colorimeter a standard curve is prepared by dissolving 0.2195 gm KH_2PO_4 in water and the solution made up to 1 litre (1ml=59 ppm phosphorus) further dilution of 10ml into 250 ml is made so as to give 1 ml= 2ppm. P Aliquots of 2, 3, 4, 5, 6, 8, 10, 15 & 20 ml of the 2 ppm solution are taken in 50 ml volumetric flasks, 2.5 ml of Barton's reagent added and the volume made up to 50 ml mark with water. After 10 minutes using a filter the color developed is read in colorimeter. A standard graph is then prepared from which Phosphorus values for experimental samples are calculated.

RESULTS AND DISCUSSION

Phosphorus is the second most important plant nutrient. It is nonrenewable resource and major plant nutrient required in sufficient amounts for higher crop. Yield as well as for proper functioning of soil biota. Phosphorous is considered one of the major constraints for successful crop production. Soil contains both organic and inorganic forms of phosphorus. The inorganic forms of the element in soil are compounds of Ca, Fe, Al and fluorine. While the organic forms are compounds of phytins, phospholipics and nucleic acids which come mainly by way of decaying vegetation. Several phosphate solubilizing micro -organization have been isolated and the very promising cultures have been identified as Pseudo-moans Striate, phosphobacterin, Aspergillus asamori, etc. shown in table. Crop Inoculants used Effect observed. Maize Phosphobacterin Increased growth, yield, and p uptake Potatoes Pseudomonas striata Increase the growth and yield. Increase the yield and Phosphorus uptake. There micro-organisms solubilize not only insoluble soil phosphates and in soluble forms of phosphatic fertilizers such as rock phosphate, but also increase the efficiency of soluble forms of phosphatic fertilizers applied to soil. Phospho-bacteria are better suited in neutral and alkaline soil and phospho fungi in acid soils. Field work has demonstrated that inoculation of plants with some P. solubilizing micro-organisms increased the concentration of available P.in the soil and enhanced the yield and phosphate uptake by the plants.

REFERENCES

[1] Gaur AC, Role of Phosphate solubilizing microorganisms and organic matter in soil productivity, Academy of Science, **1972**, 259-68.
[2] Gaur AC, Phosphate solubilizing microorganisms, Omega scientific publishes New Delhi, **1990**.
[3] Gaur AC, Gaind S, *sci. Cult* 49:, **1987**, 210-212.
[4] Rao S, Sink MK, *Can J. Microbial*. 5, **1963,** 79-85.
[5] Johnson HW, *N.Z.J. Sci*. 2, **1959**, 215-218.