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# Isolation of Phosphate Solubilizing Fungi from the Rhizospheric Soil of Wheat plant in Raipur

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**ABSTRACT**

Phosphate-solubilizing fungi were discovered in the rhizosphere soil of wheat plants in the Raipur district of India's Chhattisgarh state. A total of 19 fungi were isolated from the rhizospheric soil of wheat plants in this investigation. On the Pikovskaya's Agar medium, 12 of the 19 fungus were found to be phosphate solubilizers. In vitro, Aspergillus species, a soil isolate, demonstrated a high capacity for phosphate solubilization. Fungal strains obtained from soil with the ability to solubilize phosphate were studied in this work, and one of the fungal strains was employed as a bioinoculent.

***Keywords: Aspergillus, Penicillium , Phosphate solubilizing fungi & Solubilization***

## I.INTRODUCTION

Improving soil fertility is one of the most common methods of agricultural production. Phosphorus (P) is one of the most important plant nutrients to increase plant production. This nutrient is limited to the soil, which is still a major challenge for agricultural farmers and landowners. (1). Phosphorus is one of the major nutrients, second only to the nitrogen needed by plants. Most of the soil phosphorus, about 95-99% is present in the form of insoluble phosphates and cannot be used by plants (2). Compared with other large nutrients, phosphorus is slow and is found in plants in many soil conditions. Although phosphorus is abundant in the soil in both organic and inorganic species, it is usually a major or major factor limiting plant growth (3).

Phosphate solubilizing fungus and bacteria are known to be active organisms in this process (4,5). Molds are important organisms in the soil that often form more soil biomass than bacteria, depending on soil depth and nutrient conditions. The fungus has been reported to have a higher solubility of insoluble phosphate than bacteria (6). A wide range of soil fungi are reported to dissolve soluble phosphorus such as Aspergillus niger and Penicillium sp. which is the most common fungus that can be solved with phosphate (7). Phosphate solubilizing microorganisms tests were performed by several researchers from the soil, mangrove and rhizosphere (8). Since a large number of Chhattisgarh region is dependent on agriculture, the current research is aimed at identifying certain types of fungi that may have high phosphate solubilization.

## II . MATERIAL AND METHOD

The present investigation was carried out in the SoS Life Sciences, Pt. Ravishankar Shukla University, Raipur, Chhattisgarh. (INDIA)

### II.1-Study Site

Raipur is located in Chhattisgarh's East Central region, at latitude 21016' N, longitude 81036' E, with an elevation of 289.5 m above mean sea level. Raipur has a sub-humid climate, with an average annual rainfall of 1489 mm, of which 90% (1348 mm) falls during the monsoon season (June to September). Only 8 mm of rain falls on average throughout the rabi season (December to February). The highest temperature varied from 24.4 to 42.6 degrees Celsius, while the lowest temperature was from 10.0 to 27.50 degrees Celsius.

### II.2-Collection of soil samples

Soil samples were taken from the rhizosphere of Wheat plantations in three distinct localities in Raipur, Chhattisgarh. The samples were gathered in polythene bags, transported to the lab, and stored in the refrigerator for processing. Separate soil samples were taken from roots, air dried at room temperature, crushed, sieved, and placed in separate polythene bags. A pH metre was used to record the pH of the samples (Elico made).

### II.3-Culture media for isolation

For the isolation and preservation of phosphate solubilizing fungus, Pikovskaya's (9) agar medium (HIMEDIA) was utilised. It was made up of (g litre-1) Dextrose 10, Calcium phosphate 5, Ammonium sulphate 0.5, Potassium chloride 0.2, Magnesium sulphate 0.1, Manganease suplhate 0.0001, Yeast extract 0.5, Ferrous sulphate 0.0001, Agar 15. The medium had a pH of 7.0 (0.2).

Potato dextrose agar (PDA, HIMEDIA) was used for the isolation maintenance of fungal cultures. It contained ($g.litre^{-1}$) potato infusion 200; Dextrose 20; Agar 15 and the pH of medium was 5.6 (± 0.2) The pH of culture media was adjusted using 1N NaOH or 1N HCl. Media were sterilized by autoclaving at 121°C for 15 min.

### II.4- Isolation

All soil samples collected are used to separate the phosphate solubilizing mold from Potato Dextrose agar medium for purification and filtration. Ten grams of soil per sample was measured aseptically and transferred to a 250 ml Erlenmeyer flask containing 90 ml of distilled pure water. 1 ml supernatant aliquots from the sample were transferred to 9 ml of 0.85 percent NaCl produced in test tubes and diluted respectively in 10-1, 10-2, 10-3 10-4, 10-5 and 10-6. One ml sample of one of these purifiers was transferred to 20 ml of Potato Dextrose agar medium before solidification (temperature 450C) and

poured into a sterile Petri container after mixing. After hardening the plates are stored in the incubator at 28 ± 2ºC for 5-7 days. Fungus colonies were repeated several times on PDA plates until pure culture looked like. Pre-segmentation cultures are therefore stored in the refrigerator after growing on the sides of the PDA and used for further studies.

**II.5- Screening**

The isolates were screened by inoculating on plates containing Pikovskaya's Agar (PKA) medium (9) amended with 0.5% tricalcium phosphate (TCP) as insoluble phosphate source and were incubated at 28±2ºC for 5 days. Fungal colonies with clear halozone around them were screened as phosphate solubilizers.

**II.6- Identification**

The fungal cultures were identified on the basis of colony characteristics and microscopic examination (10, 11, and 12). Some of the fungal isolates have been sent and deposited to NFCCI for identification

**II.7-Solubilization Index (SI):**

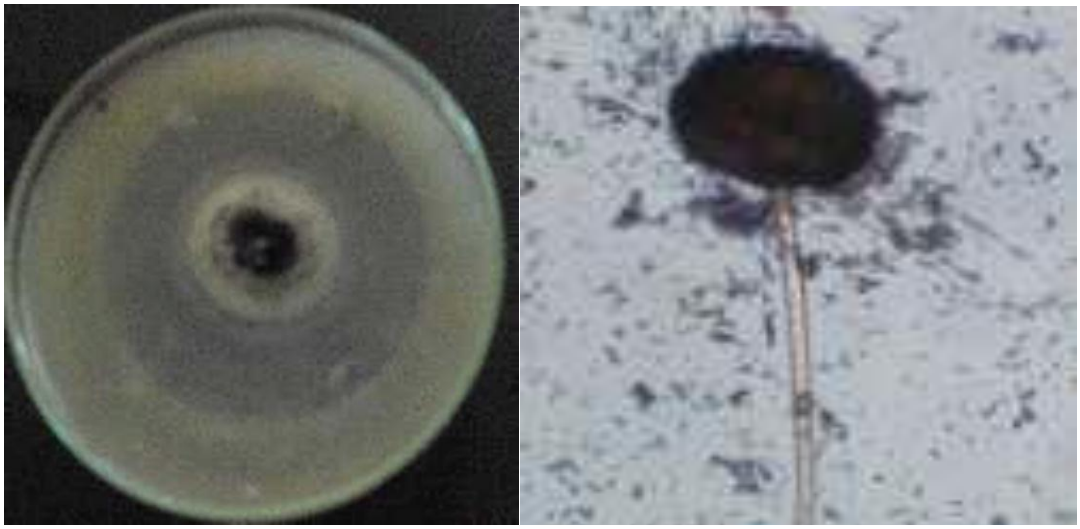
Suspensions were prepared in sterile saline (0.85 per cent NaCl) from isolate fungal cultures. Optical density of each culture was adjusted at 0.3 using colorimeter (Elico CL 157) at wavelength 520 nm. The 10 µl suspension of each isolate was placed on Pikovskaya's agar plate and incubated at 28±2ºC for 5days in incubator. Solubilization index (SI) was measured using the following formula [13].

$$SI = \frac{\text{Colony diameter + Halo zone diameter}}{\text{Colony diameter}}$$

**III.RESULTS AND DISCUSSIONS**

On the basis of the formation of transparent halozone on Pikovskaya's agar medium, 19 fungi were isolated from rhizospheric soils of chickpea plantations, and 12 were tested as phosphate solubilizers (Table). Fungi with halo zones around the colony are effective phosphate solubilizers and mostly belong to the Aspergillus and Penicillium genera. The major species was Aspergillus niger, followed by Penicillium sp. and other Aspergillus species. Mahamuni et al. (14) and Deepa et al. (13) both cited comparable findings (15).

## IV.PHOTOGRAPH OF HALOZONE FORMATION & *ASPERGILLUS NIGER*



## V.TABLE-1

| S. No | Village Name (Raipur) | Soil $p^H$ | Isolated Fungi name | Formation of Halozone |
|---|---|---|---|---|
| 01 | Jora Village | 7.70 | *Aspergillus* sp.JW1 | Yes |
| | | | *Aspergillus niger* | Yes |
| | | | *Aspergillus fumigatus* | Yes |
| | | | *Penicillium* sp.JW1 | No |
| | | | *Aspcergillus sp.JW2* | Yes |
| | | | *Rhizopus sp.JW* | No |
| | | | *Fusarium sp.JW* | No |
| | | | *Penicillium* sp. JW2 | Yes |
| 02 | Tekari | 6.8 | *Curvularia sp.TW* | No |
| | | | *Aspergillus* sp. TW | Yes |
| | | | *Fusarium spTW* | No |
| | | | *Aspergillus niger* | Yes |
| | | | *Penicillium* sp.TW | Yes |
| 03 | Nakti | 7.2 | *Aspergillus niger* | Yes |
| | | | *Aspergillus* sp.NW | Yes |

| | | | | |
|---|---|---|---|---|
| | | | *Penicillium* sp.NW | Yes |
| | | | *Curvularia sp.NW* | No |
| | | | *Mucor sp.NW* | No |
| | | | *Alternaria spNW* | Yes |

**Table: Screening of the phosphate solublizing properties by the isolated fungi on the basis of halo zone formation**.

**Qualitative assay:** The solubilization indices of different isolates ranged from 1.10 to 1.49 (**Table 2**). Fungal strains isolated from sugarcane and sugar beet rhizosphere showed SI in range of 1.13 to 1.59 (14). Alam et al. (16) reported SI of the fungal strains isolated from maize rhizosphere that ranged from 1.53 to 1.80.

**TABLE-II**

| **S. No** | **Name of fungi** | **Solubilization Index (SI)** | | |
|---|---|---|---|---|
| | | **A(mm)** | **B(mm)** | **SI** |
| 1 | *Aspergillus* sp.JW1 | 49 | 33 | 1.48 |
| 2 | *Aspergillus niger* | 50 | 35 | 1.42 |
| 3 | *Aspergillus fumigatus* | 52 | 37 | 1.40 |
| 4 | *Aspcergillus sp.JW2* | 50.5 | 33.8 | 1.49 |
| 5 | *Penicillium* sp. JW2 | 42 | 31 | 1.35 |
| 6 | *Aspergillus* sp. TW | 78 | 69 | 1.13 |
| 7 | *Aspergillus niger* | 44 | 39.9 | 1.10 |
| 8 | *Penicillium* sp.TW | 43 | 29.9 | 1.43 |
| 9 | *Aspergillus niger* | 51 | 36 | 1.41 |
| 10 | *Aspergillus* sp.NW | 58 | 45 | 1.28 |
| 11 | *Penicillium* sp.NW | 44 | 31.3 | 1.40 |
| 12 | *Alternaria sp.NW* | 40 | 32 | 1.25 |

A= (Halozone + Colony) diameter, B= Colony Diameter.

## VI.CONCLUSION

According to the findings, the wheat rhizosphere contains a variety of phosphate-solubilizing fungus, with Aspergillus and Penicillium being the most prevalent forms. Phosphate solubilization was highest in Aspergillus species, suggesting that it might be employed as a phosphate biofertilizer for wheat and other agricultural plants. To establish its inoculation effects on various agricultural plants, further nursery and field studies are needed.

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