Evaluation of different carrier material for survival *Aspergillus niger* a phosphate solubilizing organism

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**Abstract**
Species of *Aspergillus* viz. *A. niger, A. flavus, A. terreus* were found to be effective phosphorous solubilizers, in order to utilize these species for large scale production of bioformulation, the basic studies on survival of the test organism in different carrier material was done. Out of six carrier materials tested for survival of *Aspergillus* spp. Lignite was found to be the most effective carrier material for all *Aspergillus* spp. with good colony growth up to 90 days.

**Keywords:** *Aspergillus* spp, carrier material, lignite, survival

**Introduction**
Optimum development of crops demands a high and often costly input of phosphate fertilizer. Current concept of sustainability involves application of alternative strategy based on the use of less expensive natural sources of plant nutrients like rock phosphate. In addition, chemical fertilizers are highly expensive and have adverse effect on soil.

A soil bacteria belong to genera pseudomonas and fungi belong to genera *Penecillium* and *Aspergillus* have ability to bring insoluble phosphate present in soil into soluble forms by secreting organic acid viz., formic, acetic, propionic, lactic, Izglycolic, fumaric succinic acid etc. these acids lower the ph and bring about the dissolution of bound forms of phosphate. Phosphate solubilizing microorganisms have been reported to occur in different environmental niches and are abundant in rhizospher (Nahtiyl et.al.2000 and Chen et.al.2002) [6, 4]. Fungi have been reported to possess greater ability to solubilize insoluble phosphate than bacteria (Nahas, 1996) [8].

Biofertilizers are microbial inoculants containing either living or latent cells of nitrogen fixing or phosphate solubilizing organisms. The technology of producing these inoculants consist of cultivating efficient strains of microbes in suitable media under controlled condition and mixing the cultural broth with sterile carrier material by following the standard procedure. Among the phosphate solubilizing fungi *Aspergillus* and *penecillium* species are widely exploited for their biofertilizer production. Phosphate solubilizing *Aspergillus* spp. are *A. niger, A. flavus, A. terreus, A. sydowii, A. amstelodami, A. versicolor, Aspergillus niger* is a versatile phosphate solubelizer and is abundantly present in different soil types (Gaur 1990). It solubilize eight insoluble metal compounds viz., Al2O3, Al2(PO4)3, CaCO3, Ca3(PO4)2, Mn (PO4)2 ZnO and Zn3(PO4)2. *Aspergillus* lowers the soil ph and brings about dissolution of immobile forms of soil phosphate. Some organic acid produced by it may chelate calcium, aluminium, ferrous and magnesium further increasing phosphorus availability.

**Material and Methods**
Cultures- Fungal cultures which are found to be good phosphate solubilizer were obtained from Biofertilizer Production center, Department of plant pathology. These fungal spp. are *Aspergillus niger, A. vercicolor and A. sydowii*. Potato dextrose agar (PDA) was used for maintaining of the cultures during the course of investigation.

**Effect of different carrier materials**
The effect of carrier material was studied for 30, 60and 90 days. Different carrier material used as a lignite, lignite + soil, lignite + sawdust, sawdust, compost and talc.
All the carrier materials were sterilized for two times at 1210°C in an autoclave. The bioinoculants in the form of sporulated broth culture of Aspergillus niger, A. flavus and A. terreus was mixed separately in 2:1 proportion and the poly bags (75 µ milky white) were filled with 250gm bio- inoculants. Bags were closed with sealing machine and kept in room temperature for further observation. The bags from each carrier containing the test inoculants were opened after 30, 60 and 90 days for studying the survival of Aspergillus spp. The serial dilution method was followed for observing CFU load in carrier material by using PDA. The inoculated plates were incubated at 200°C for 48hrs to record the number of colonies appearing in each petri plates. Three replications were maintained for each carrier and the Aspergillus spp.

### Effect of carrier material

#### Table 1: CFU after 30 days (in numbers)

| Carrier material          | A. niger* | A. flavus* | A. terreus* | Mean |
|---------------------------|-----------|------------|-------------|------|
| Lignite                   | 28.66     | 26.00      | 24.66       | 26.44|
| Lignite+soil(1:1)         | 24.66     | 24.66      | 24.00       | 24.44|
| Lignite+sawdust(1:1)      | 21.33     | 22.33      | 21.66       | 21.77|
| Sawdust                   | 3.00      | 3.67       | 3.66        | 3.44 |
| Compost                   | 19.66     | 16.00      | 15.66       | 17.10|
| Talc                      | 2.33      | 18.33      | 19.67       | 19.77|
| Mean                      | 19.77     | 18.49      | 18.22       |      |
| S.E.m±                    | 0.25      |            | 0.95        |      |
| Aspergillus spp           | 0.18      |            | 0.67        |      |
| Carrier material x Aspergillus spp | 0.43 |            | 1.65        |      |

*Means of three replications

### Effect of carrier material on survival

It is revealed from table 1 that lignite was the best carrier material giving highest colony count (26.44) after 30 days of storage. It was followed by lignite + soil with 24.44 colony count Lignite + sawdust ranked third where colony count was 21.77. In sawdust colony count was 19.7 while lowest colony count (3.44) was obtained in sawdust.

### Effect of Aspergillus spp

Table one shows that highest colony count was recorded by A. niger (19.77) followed by A. flavus and A. terreus where colony count was 18.49 and 18.22 respectively and was on par with each other.

### Effect of interaction of month and carrier material

Highest colony count was found in interaction between lignite and A. niger (28.66) and lowest colony count of A. terreus was found in sawdust (3.00).

#### Table 2: CFU after 60 days (in numbers)

| Carrier material          | A. niger* | A. flavus* | A. terreus* | Mean |
|---------------------------|-----------|------------|-------------|------|
| Lignite                   | 24.66     | 24.00      | 22.66       | 23.77|
| Lignite + soil (1:1)      | 23.00     | 23.00      | 21.00       | 22.33|
| Lignite + sawdust (1:1)   | 20.33     | 21.00      | 19.33       | 20.22|
| Sawdust                   | 3.00      | 3.67       | 3.33        | 3.66 |
| Compost                   | 14.33     | 14.33      | 15.66       | 14.77|
| Talc                      | 16.66     | 17.00      | 17.66       | 16.91|
| Mean                      | 16.99     | 17.16      | 16.06       |      |
| S.E.m±                    | 0.21      |            | 0.80        |      |
| Aspergillus spp           | 0.15      |            | 0.56        |      |
| Carrier material x Aspergillus spp | 0.36 |            | 1.38        |      |

*Mean of three replications

### Effect of carrier material

It is clear from the table tow that highest colony count was obtained when lignite was used as carrier material irrespective of the Aspergillus spp. The colony count in this treatment was 23.77. It was followed by lignite + soil which recorded 22.33 colonies irrespective of Aspergillus spp. The colony count in lignite + sawdust was 20.22 irrespective of Aspergillus spp. Sawdust was found to be poor carrier material for Aspergillus spp. as it recorded least colony count (3.66).

### Effect of Aspergillus spp

It was revealed that, A. flavus was superior in colony development as a maximum colonies (17.16) were obtained irrespective of nitrogen source used. It was followed by A. niger and A. terreus which are at par.

### Interaction of Aspergillus spp and carrier material

The interaction effect of A. niger and carrier material depicted table two revealed that the interaction between A. niger and lignite was highly significant as it recorded the maximum colony count (24.66). It was followed by the interaction between A. flavus and lignite which recorded the colony count to the tune of (24.00). The interaction between A. niger and sawdust was highly undesirable as 3.00 colonies were observed.
between lignite and sawdust was highly undesirable as 3.00 colonies were observed after 90 days these results were in accordance with results reported by Kandasmy and Prasad (1971) who observed that lignite is a good source of carrier material irrespective of the Aspergillus spp grown. The colony count in this treatment was 15.44. It was followed by lignite + soil which recorded 14.55 colonies irrespective of Aspergillus spp. and on at par with talc. Colony count in lignite + sawdust was 13.66. Sawdust was found to be poor carrier material for development of Aspergillus spp. as it recorded least colony count (3.22).

It is revealed that, A. niger was superior in colony development as a maximum colonies (12.77) were obtained irrespective of nitrogen source used. It was on par with A. terreus.

### Interaction of Aspergillus spp and carrier material

The interaction effect of the A. niger and carrier material depicted in table three revealed that the interaction between A. niger and lignite was highly significant as it is recorded the maximum colony count (17). It was followed by the interaction between lignite + soil and A. niger which recorded the colony count to the tune of 15.66. The interaction between A. terreus, A. niger and sawdust was highly undesirable as only 3.00 colonies were observed after 90 days.

### Discussion

#### Effect of carrier material

Maximum colony count was obtained when lignite was used as a carrier material irrespective of the Aspergillus spp grown after 90 days. The colony count in this treatment was 15.44. It was followed by lignite + soil which recorded 14.55 colonies irrespective of Aspergillus spp. and on at par with talc the colony count in the lignite + sawdust treatment was 13.66. Sawdust was found to be poor carrier material for development of Aspergillus spp. as it recorded least colony count (3.22). The interaction between A. niger and lignite was highly significant as it recorded the maximum colony count (17.00). It was followed by the interaction between lignite + soil and A. niger which recorded the colony count to the tune of 15.66. The interaction between A. terreus, A. niger and sawdust was highly undesirable as 3.00 colonies were observed after 90 days these results were in accordance with results reported by Kandasmy and Prasad (1971) who observed that lignite is a good source of carrier material for preparation of microbial inoculants. Similarly Menaka and Alagwadi (2007) used 200 mesh lignite as a carrier material for phosphate solubilizing bacteria. Similar results were obtained by Heijen et al. (1993) also found lignite and talc are effective carriers for phosphate solubilizing Aspergillus spp.

### Summary and conclusion

Lignite was found to be good carrier material for all Aspergillus spp. They showed good colony growth up to 90 days but colony number was reduced compared to 30 and 60 days. A. niger showed good response in lignite after 30, 60 and 90 days with colony count 28.66, 24.66 and 17.00 respectively. It was followed by lignite + soil with colony count 24.66, 23, and 15.66 after 30, 60 and 90 days respectively.

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### Table 3: CFU after 90 days (in numbers)

| Carrier material | A. niger* | A. flavus* | A. terreus* | Mean |
|------------------|----------|------------|------------|------|
| Lignite          | 17.00    | 14.33      | 15.00      | 15.44|
| Lignite + soil   | 15.66    | 13         | 15.00      | 14.55|
| Lignite + sawdust| 13.66    | 13.66      | 13.66      | 13.66|
| Sawdust          | 3.00     | 3.67       | 3.00       | 3.22 |
| Compost          | 12.00    | 8.66       | 12.33      | 10.99|
| Talc             | 15.33    | 13.33      | 15.33      | 14.66|
| Mean             | 12.77    | 11.10      | 12.38      |      |

S.E.m± C.D. carriers material

| Carrier material | 0.18 | 0.13 | 0.32 |
|------------------|------|------|------|
| Mean             | 0.71 | 0.50 | 1.23 |

*Mean of three replications