

IN-VITRO STUDIES ON COMPATIBILITY BEHAVIOR AMONG MICROBIAL INOCULANTS

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Abstract: The present study was conducted during 2017-19 at Department of Plant Pathology and Agricultural Microbiology, Post Graduate Institute, M.P.K.V., Rahuri-413722. All the microbes have to prove effective in colonization of the plant roots for efficient function under natural soil conditions. Compatibility between the PGPR microbes to colonize the root system without inhibiting each other is a pre-requisite for success of using multiple microbes in a crop field. In our study, among the different microbes tested for their compatibility in culture growth, the all species of bioinoculants were found to compatible the growth of other species as evident from the no zone of inhibition observed in the plates. All other microbes viz; *Trichoderma viride*, *Pseudomonas fluorescens*, *Azotobacter chroococcum* and *Bacillus polymyxa* on NA media were compatible with each other. These results have made us to choose the best two candidate bacteria for further studies.

Keyword: Bioinoculants, Compatible, PGPR microbes

INTRODUCTION

The mixed culture approach is simply an effort to apply these principles to natural systems such as agricultural soils, and to shift the microbiological equilibrium in favour of increased plant growth, production and protection. The use of mixed cultures of beneficial microorganisms as soil inoculants is based on the principles of natural ecosystems which are sustained by their constituents; that is, by the quality and quantity of their inhabitants and specific ecological parameters, i.e., the greater the diversity and number of the inhabitants, the higher the order of their interaction and the more stable the ecosystem (Higa 1994). Compatibility of the inoculants *Rhizobium* sp, *Bacillus megaterium* and *Pseudomonas fluorescens* through cross streak assay and found that inoculates were compatible with each other and hence able to grow simultaneously without any inhibition in growth (Anandraj and Delapierre 2010).

Garikapathi Sateesh and Sivasakthivelan (2013) studied the compatibility behaviour among the bioinoculant. This study clearly indicated that the effect of bio-inoculation with consortium viz., *Trichoderma viride* + *Pseudomonas fluorescens* + *Azotobacter chroococcum* was much better when compared to dual and single inoculation which enhanced the growth and yield parameters when compared to control thereby improving the soil health. The present study was under taken to assess the effect of the mixed inoculants of *Trichoderma viride*, *Pseudomonas fluorescens* and *Azotobacter chroococcum* on the growth, soil fertility and yield of chillies. The compatibility of the inoculants *Trichoderma viride*, *Pseudomonas fluorescens* and

Azotobacter chroococcum was tested through cross streak plate assay. The inoculants were found to be compatible with each other and were able to grow simultaneously without any inhibition in growth.

MATERIALS AND METHODS

Source of Microbial Inoculant

The Microbial inoculant viz; *Trichoderma viride*, *Pseudomonas fluorescens*, *Azotobacter chroococcum* and *Bacillus polymyxa* (PSB) were obtained from Department of Plant Pathology and Agricultural Microbiology, Mahatma Phule Krushi Vidyapeeth, Rahuri.

Maintenance of Pure Culture

The bioagents viz; *Trichoderma viride* and *Pseudomonas fluorescens* were sub-cultured on PDA (Peeled potato 200g, Dextrose 20g, Agar 15g), TSM (Glucose 3.0 g, Di-potassium hydrogen orthophosphate 0.9 g, Ammonium Nitrate 1.0 g, Potassium chloride 0.15 g, Magnesium sulphate 0.2 g, Agar 15 g, Rose Bengal 0.15 g, Chloramphenicol 0.25 g, Metaxyl 0.30 g) and Kings B (Proteous peptone 20 g, Di-potassium hydrogen orthophosphate (K₂HPO₄) 2.5 g, Glycerol 15 ml, Magnesium sulphate (MgSO₄ 7H₂O) 6 g, Agar 15 g) medium slants, respectively. Similarly, the biofertilizers viz; *Azotobacter chroococcum* and *Bacillus polymyxa* were sub-cultured on Jensen's medium (Sucrose 20 g, Di-potassium hydrogen phosphate 1g, Magnesium sulphate 0.5g, Sodium chloride 0.5g, Ferrous sulphate 0.1 g, Na₂MoO₄ 0.005g, Calcium carbonate 2 g, Agar 15g) and Pikovaskiya's medium slants (Glucose 10g, Tricalcium phosphate 5.0g, Ammonium sulphate 0.5 g, Potassium chloride 0.2 g, Magnesium sulphate 0.1g, Manganese sulphate Trace, Ferrous

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sulphate Trace, Yeast extract 0.5 g, Agar 15 g.), respectively. Then incubated the inoculated slants at $27 \pm 2^\circ\text{C}$ for 7 days for sufficient growth. The slants were stored in refrigerator at 4°C and subculture periodically.

Compatibility

The dual culture technique were used to see the compatibility activity between bioinoculants viz; *Trichoderma viride*, *Pseudomonas fluorescens*, *Azotobacter chroococcum* and *Bacillus polymyxa* (PSB) on NA media. The streaking was made each of microbial strain in combination with another strain, then incubated the inoculated plates at $27 \pm 2^\circ\text{C}$ for 3-5 days for sufficient growth and observed for any growth restriction with formation of inhibition zone. The results were interpreted on the basis of positive (++) means doesn't seen any inhibition zone and negative (--) means formation of inhibition zone in dual culture.

RESULTS AND DISCUSSION

Compatibility Behaviour of Microbial Inoculants

Experiment was carried out to see the synergetic or antagonistic activity between bioinoculants used viz; *Trichoderma viride*, *Pseudomonas fluorescens*, *Azotobacter chroococcum* and Phosphate solubilizing bacteria (*Bacillus polymyxa*) on NA media. Results were represented in Table: 4.1 and plate: 1

Results showed that all the bioinoculants used were compatible with each other and dose not showed any antagonistic effect on another used bioinoculant. The *Azotobacter chroococcum* and *Bacillus polymyxa*, *Bacillus polymyxa* and *Pseudomonas fluorescens* were highly compatible with each other as compare to other bioinoculants.

Table 1. Compatibility study among bioinoculants

Microbial inoculants	<i>T. viride</i>	<i>P. fluorescens</i>	<i>A. chroococcum</i>	<i>B. polymyxa</i>
<i>T. viride</i>	++	++	++	++
<i>P. fluorescens</i>	++	++	++	++
<i>A. chroococcum</i>	++	++	++	++
<i>B. polymyxa</i>	++	++	++	++

++ indicates Compatible, whereas – indicates Non-compatible

The results of present investigation correlates with the results of earlier research workers like Manjula et al. (2004) reported in vitro compatibility of *Pseudomonas fluorescens* and *Trichoderma sp.* in dual culture and found that *Pseudomonas fluorescens* had no effect on growth of *Trichoderma sp.* or vice-versa. Bagwan (2010) reported that among the botanicals tested, the interesting thing observed that, neem oil (5%), neem leaves extract (10%), wild sorghum leaves extract (10%), neem cake, castor cake and mustard cake extract (10%) enhanced the growth of *Trichoderma*. This finding indicates that applications of *Trichoderma* would be compatible with neem oil, neem leaves extract, wild sorghum leaves extract, neem cake, castor cake and mustard cake extracts for the integrated management of soil borne diseases of groundnut. Kumar Rakesh (2010) reported that *Trichoderma* is compatible with biofertilizers like *Rhizobium*, *Azospirillum*, *Bacillus*

subtilis and phosphor-bacteria.

The compatibility studies revealed the isolates of *T. viride* (Tv1), *P. fluorescens* (Pf1) and *B. subtilis* (Bs16) were compatible with each other and also with organic amendments and micronutrients (Latha et al. 2011). Garikapati Sateesh et al. (2013) studied the compatibility behaviour among the bioinoculant. The compatibility of the inoculants *Trichoderma viride*, *Pseudomonas fluorescens* and *Azotobacter chroococcum* was tested through cross streak plate assay. The inoculants were found to be compatible with each other and were able to grow simultaneously without any inhibition in growth. Madewar et al. (2013) were concluded that, among five *Trichoderma spp.* tested with *Bacillus polymyxa*, *T. viride* was found most compatible with *Bacillus polymyxa* followed by *T. harzianum*, *T. hamatum* and *T. longibrachiatum*

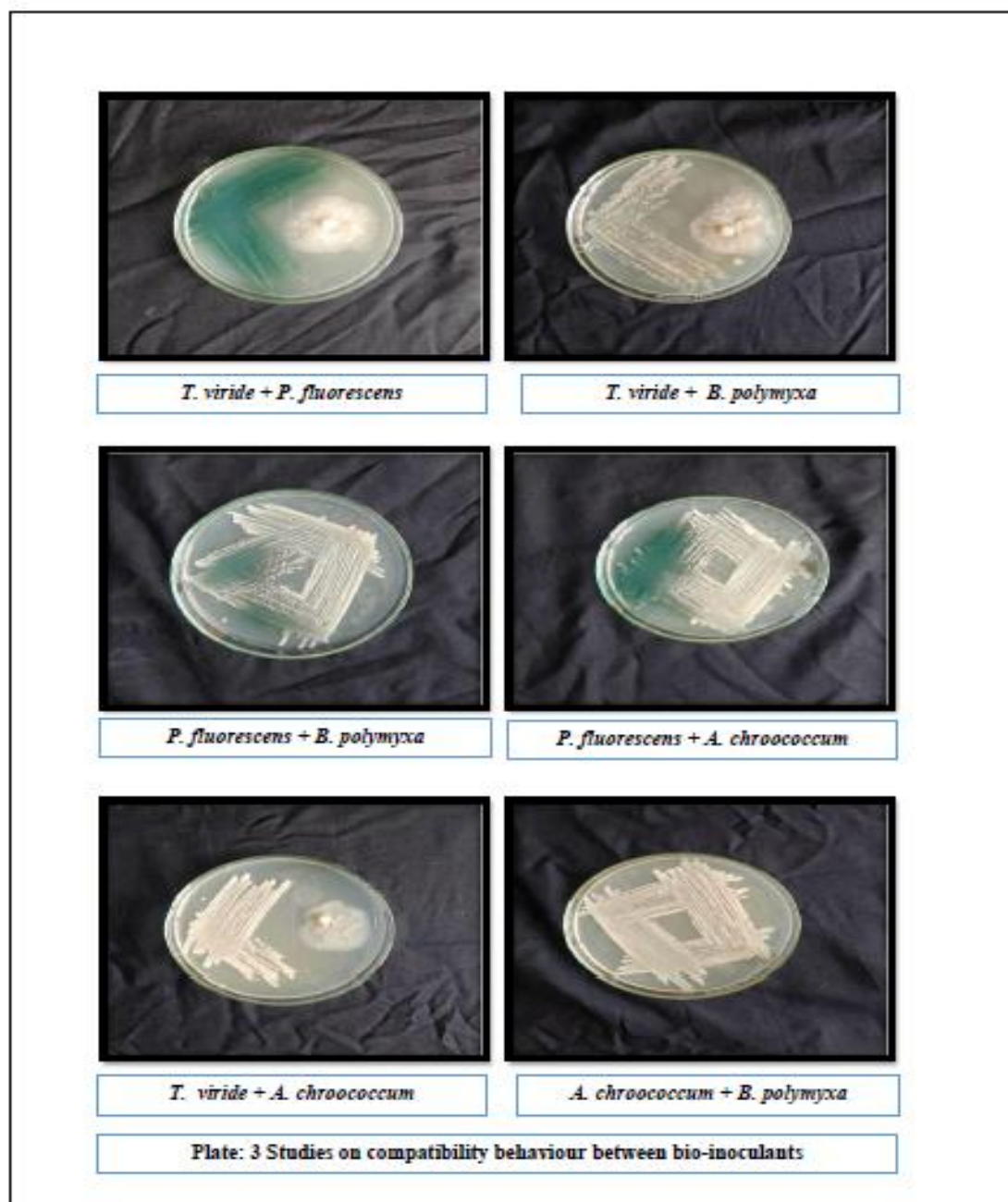


Plate: 1. Studies on compatibility behavior among bioinoculants.

SUMMARY AND CONCLUSIONS

Experiment was carried out to see the synergetic or antagonistic activity between bioinoculants used viz., *Trichoderma viride*, *Pseudomonas fluorescens*, *Azotobacter chroococcum* and *Bacillus polymyxa* on NA media. Results showed that all the bioinoculants used were compatible with each other and not showed any antagonistic effect. The *Azotobacter chroococcum* and *Bacillus polymyxa*, *Bacillus polymyxa* and *Pseudomonas fluorescens* were highly compatible with each other as compared to other bioinoculants.

REFERENCES

- Anandaraj, B. and Leema Rose Delapierre** (2010). A studies on influence of bio inoculants (*Pseudomonas fluorescens*, *Rhizobium* sp., *Bacillus megaterium*) in green gram. *J. Bio. Sci. Tech.*, 1(2): 95-99.
- Bagwan, N. B.** (2010). Evaluation of *Trichoderma* compatibility with fungicides, pesticides, organic cakes and botanicals for integrated management of soil borne diseases of soybean [*Glycine max* (L.) Merrill]. *International Journal of Plant Protection*, 3(2): 206-209.

Garikapati, Sateesh and Sivasakthivelan, P. (2013). Studies on the influence of bioinoculant consortium on chillies and its effects on soil health management, *Int.J.Chem.Tech. Res.* 5, pp3.

Higa, T. (1994). Effective microorganisms: A new dimension for nature Farming. p. 20-22. In: Parr JF, Hornick SB and Simpson ME (Eds), Proceedings of the Second International Conference on Kyusei Nature Farming. US Department of Agriculture, Washington DC, USA.

Kumar, Rakesh, Kumar, Rameshand Kumar, Prabha (2010). Effect of integrated use of chemical fertilizers, biofertilizers and biostimulants in gladiolus (*Gladiolus grandiflorus* L.) cv. Sancerre Progressive horticulture, *Indian journals*, 43(1): 149-152.

Latha, P., Anand, T., Prakasam, V., Jonathan, E. I., Paramathma, M. and Samiyappan, R. (2011). Combining *Pseudomonas*, *Bacillus* and *Trichoderma* strains with organic amendments and micronutrient to enhance suppression of collar and root rot disease in physic nut. *Applied Soil Ecology*, 49, pp 215-223

Madewar Ganesh Prabhakar (2013). Compatibility studies on Phosphate Solubilizing Microorganisms with *Trichoderma spp.* M.Sc. Thesis, MPKV, Rahuri.

Manjula, K., Kishore, G.K., Girish, A.G. and Singh, S.D. (2004). Combined application of *Pseudomonas fluorescens* and *Trichoderma viride* has an improved biocontrol activity against stem rot in groundnut. *Pl. Pathol. J.*, 20(1): 75-80.