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EFFECT OF *RHIZOBIUM* Spp., ON *MACROPHOMINA PHASEOLINA*

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In dual culture plate assays, indigenous *Rhizobium* strains isolated from nodules obtained from the fields of Karachi inhibited radial growth of *Macrophomina phaseolina, Rhizobium leguminosarum, R. meliloti* and *R. japonicum* causing growth inhibition. In *vitro* also led to a significant reduction in the severity of *Macrophomina* root rot of mungbean, okra and sunflower in green house experiments. These data suggest that a potential exists for reducing *Macrophomina* root rot by employing nodulating of *Rhizobium* strains which are highly antagonistic to *M. phaseolina.*

Key words: Seed bacterization, Biological control, *Rhizobium-Macrophomina* interaction.

INTRODUCTION

Macrophomina phaseolina (Tassi) Goid. causes seedling blight, root rot, stem rot and pod rot of more than 500 species of plants [7]. The fungus is widely distributed in tropical and subtropical countries of the world, of which at least 40 hosts have been recorded from Pakistan alone [1]. Apart from the use of chemical fungicides, biological control agents have also been found to protect seeds and control root diseases [2,3,4,5]. Delivery of antagonists directly to soil requires a large amount of material; seed treatment is therefore an attractive method for introducing biological control agents into the soil-plant environment. *Rhizobium* spp. is used on seed to induce nodulation and increase soil fertility. Experiments were, therefore, carried out to study the effect of *Rhizobium* spp., on Macrophomina phaseolina.

MATERIAL AND METHOD

M. phaseolina (K.U.M. H. Cult. 54) isolated from root rot of cotton was used. The fungus was multiplied on cornmeal sand medium for 2 months at 30°. The sclerotia separated through 100 μm sieve were used to artificially infest soil @ 30 scl g^{-1} soil. Soil used was sandy loam, pH 7.5, with a natural population of 4 sclerotia g^{-1} soil.

Cultures of *Rhizobium leguminosarum* (KUMH Cult. 371), *R. japonicum* (Cult. 373) and *R. meliloti* (Cult. 372) isolated from nodules of pea, lucerne and soybean from the fields of Karachi were grown on PDA at 30° for 3 days. Bacterial cells were harvested by scraping the culture surface with a spatula and the seeds of mungbean, okra and sunflower dipped in cell suspenion using 1 % gum arabic as a sticker. The seeds were allowed to dry before they were sown in 15 cm diam, plastic pots containing 200 g soil artificially infested with *M. phaseolina.* There were 10 seeds per pot with 4 replicates of each treatment. Soil was adjusted and maintained at 50 % MHC by the daily addition of water. Seedlings were uprooted after 10 days, washed in running tap water, surface disinfected with 5 % $CaOCl_2$ and 1 cm pieces transferred on PDA containing penicillin and streptomycin. Dishes were incubated for 4 days at 28° to see *Macrophomina* infection.

RESULTS AND DISCUSSION

Radial growth of *M. phaseolina* was inhibited when grown opposite *R. leguminosarum, R. japonicum* and

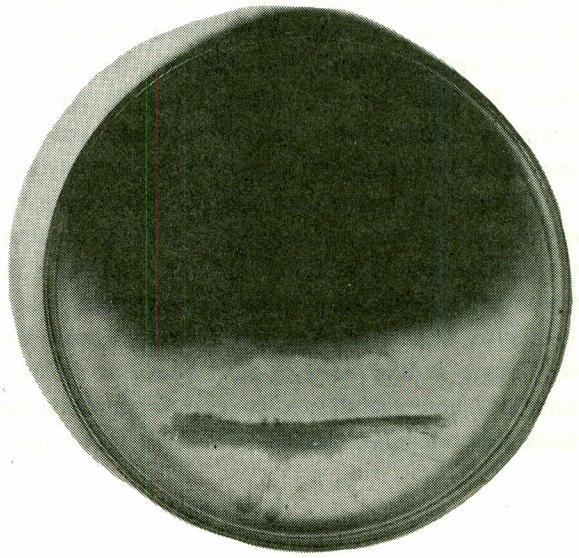

Fig. 1. Inhibition of *Macrophomina phaseolina* (top of the plate) by *Rhizobium meliloti* (bottom of the plate) on Czapek Dox agar.

Table 1. Effect of seed bacterization on *Macrophomina* infection of mungbean, okra and sunflower seedlings in naturally and artificially infested soil at 50 % MHC after 10-days.

Treatments	No. of Sci. g^{-1} soil		Colonization %		
			Mungbean	okra	Sunflower
Control	N.I.S	4	14	10	6
	A.I.S	30	28	16	21
Rhizobium lequminosarum	NIS	4	3 (-76%)	0 (-100%)	0 (-100%)
	AIS	30	12 (-55%)	0 (-10%)	0 (-44%)
R.japonicum	NIS	4	12 (-10%)	0 (-100%)	0 (-100%)
	AIS	30	11 (-59%)	9 (-45%)	13 (-36%)
R.meliloti	NIS	4	12 (-14%)	10 (0%)	0 (-100%)
	AIS	30	8 (-71%)	20 (+20%)	24 (+14%)

Figures in parentheses are % reduction (–) or increase (+) over control.
NIS = Naturally infested soil
AIS = Artificially infested soil

R. meliloti producing a zone of inhibition of 20-25 mm. (Fig. 1). In soil, strains of *Rhizobium* also caused significant reduction in the severity of *Macrophomina* infection of mungbean, okra and sunflower. With a low inoculum level of *M. phaseolina* sclerotia in soil, upto 100 % control of Macrophomina infection was observed in sunflower and okra seedlings. Such similar reports have been made by Purkayastha *et al.* [6] who found that an interaction of *Rhizobium* and *Macrophomina* reduced charcoal rot in soybean. Similarly Tu [8] observed that *R. japonicum* significiantly reduced root rot of soybean caused by *Phytophthora meqasperma* f. sp. *glycine*. Presumably bacterial colonization in hyphal tips prevented the fungus from coming in contact with host cells. Our study demonstrated that *M. phaseolina* is inhibited by *Rhizobium* spp., and that a potential exists for reducing *Macrophomina* root rot by employing nodulating *Rhizobium* strains as seed treatment. There is also need to asses the root colonizing capacity of microorganisms after seed colonization since portions near inoculum sources are likely to be heavily colonized than portions farther away.

REFERENCES

1. A. Kafi, A. Ghaffar and R. Mirza,. Pakistan J. Sci. Ind. Res., 7, 71 (1964).
2. G.E. Harman, I. Chet and R. Baker, Phytopathol., **71**, 569 (1981).
3. Y. Henis, A. Ghaffar and R. Baker, Phytopathol.,**68**, 900 (1978).
4. T. Kommedahi and C.E. Windels "Biological Control in Crop Production". ed. G.C. Papavizas, Allanheld (Osmum & Co., Totiwa, N.J., 1981), pp. 227-48.
5. J.J. Marios, D.J. Mitchell and R.M. Sonoda, Phytopathol. **71**, 1257 (1981).
6. R.P. Purkayastha, U. Menon and B.N Chakraborty, Indian J. Exp. Biol., **19**, 462 (1981).
7. J.B. Sinclair, Phytopathol. Soc., 104 pp. (1982).
8. J.C. Tu, Physiol. Plant Pathol., **12**, 233 (1978).