Biocontrol of pea root rot incited by *Fusarium solani* f. sp. *pisi* with rhizosphere mycoflora

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ABSTRACT: The root rot disease of pea (*Pisum sativum* L.) incited by *Fusarium solani* f.sp. *pisi* cause considerable crop losses in northern India. The fungal isolates of pea rhizosphere were evaluated for their biocontrol potential against *Fusarium solani* f.sp. *pisi* under *in vitro* study (dual culture method) and sick soil (pot) condition. Under *in vitro* study, isolate *Trichoderma viride* showed the strongest antagonistic activity towards *F. solani* f.sp. *pisi* in dual culture followed by *Aspergillus niger, A. terreus, A. sydowi, A. flavus* and *Spicaria sylvatica*. The two highly efficient antagonists- *T. viride* and *A. niger* were studied individually and in combinations in relation to the biocontrol of pea root rot under pathogen sick soil (pot) condition. Both the antagonists controlled the disease more efficiently when used individually than those used in combinations. *T. viride* and *A. niger* applied individually @15 gm/5kg sick soil recorded the disease severity of 33.3% and 40% respectively, while the disease severity under untreated sick soil was 96.6%.

Key words : Pea, *Fusarium solani* f.sp. *pisi*, biocontrol, antagonist

The root rot disease incited by *Fusarium solani* (Martius) Appel and Wolleweber f. sp. *pisi* (John) Snyder and Hansen is a serious disease of pea (*Pisum sativum* L.) throughout the world (Furgel, 1984; Kraft et al. (1998). This disease is soil borne and causes considerable crop losses in northern India. as high as 97 per cent (Sen and Mazumdar, 1974).

In spite of their effectiveness, chemicals are not advocated for controlling the soil borne plant pathogens in view of their prohibitive cost and threat to beneficial soil microflora, hence the interest is shifting towards biological control. The rhizosphere microflora are ideal as biocontrol agents because the rhizosphere provides the first line of defence to root surface against invading pathogen (Weller, 1988).

In the present investigation, the biocontrol potential of pea rhizosphere mycoflora has been studied in relation to their effect on the development of root rot incited by *Fusarium solani* f. sp. *pisi*.

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**MATERIALS AND METHODS**

**Isolation of rhizosphere mycoflora**

The rhizosphere soils were collected from healthy and root rot affected pea plants starting from 10 days after emergence of seedlings upto 60 days at 10 days intervals. The serial dilutions (10^-3) were prepared from one gram of each rhizosphere soil and 1 ml of each dilution was plated on Martin’s Rose Bengal Agar medium and incubated at 30°C for 7 days. Each fungal colony was transferred to PDA slant and purified by hyphal tip method. The fungi, which appeared frequently, were identified based on morphological and cultural characters, as *Aspergillus niger, A. flavus, A. terreus, A. sydowi, Trichoderma viride, Spicaria sylvatica, Rhizopus nigricans* and *Fusarium sp*.

**Isolation and pathogenicity of the pathogen**

The fungus *Fusarium solani* f. sp. *pisi* was frequently isolated from the root rot affected pea seedlings and the pathogenicity was proved by the method described by sagar and sugha (1997).
Screening of rhizosphere mycoflora for antagonistic activity against the pathogen

The mycoflora, isolated from pea rhizosphere, were screened for their antagonistic activity towards the target pathogen by dual culture method (Martyn and Stack, 1990). All the rhizosphere isolates and the pathogen were multiplied in PDA plates for ten days. A 5 mm disc of each rhizosphere fungus was inoculated at one side of PDA plate aseptically and a 5 mm disc of the pathogen was also inoculated at the opposite side of the same plate in such a way that both the inocula were 4 cm apart. The plates were then incubated at 30°C for 7 days and the inhibition of the growth of the pathogen was recorded. Based on dual culture study, the mycoflora Trichoderma viride, Aspergillus niger, A. terreus, A. sydowi and A. flavus, Spicaria sy/vatica, showed antagonistic activity towards the pathogen. Two highly efficient strains of antagonists viz. A. niger and T. viride were further studied in relation to their effect on the development of pea root rot caused by Fusarium solani f. sp. pisi under screen house condition.

Effect of antagonists on the disease development

Two highly efficient strains of antagonists Aspergillus niger and Trichoderma viride were multiplied on sand maize meal (9:1) medium for 15 days. Similarly the pathogen was also multiplied on the same medium for 15 days. The sickpots were prepared by using the inoculum of the pathogen @ 30 gm/kg soil. Each pot contained 5 kg sick soil. The pots were left for 48 hrs for stabilization before starting the experiment. Thereafter the sick pots were inoculated with the biomass of Aspergillus niger and Trichoderma viride as per treatment given below:

Treatments (gm/pot)

**Aspergillus niger (An)**
1. An -  5 gm
2. An -  10 gm
3. An -  15 gm

**Trichoderma viride (TV)**
4. TV -  5 gm
5. TV -  10 gm
6. TV -  15 gm

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Treatments (gm/pot)</th>
<th>Disease severity (%)</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>Aspergillus niger (An)</td>
<td></td>
</tr>
<tr>
<td>1.</td>
<td>An (5 gm)</td>
<td>77.00 (61.35)</td>
</tr>
<tr>
<td>2.</td>
<td>An (10 gm)</td>
<td>43.30 (41.15)</td>
</tr>
<tr>
<td>3.</td>
<td>An (15 gm)</td>
<td>40.00 (39.23)</td>
</tr>
<tr>
<td></td>
<td>Trichoderma viride (TV)</td>
<td></td>
</tr>
<tr>
<td>4.</td>
<td>TV (5 gm)</td>
<td>46.60 (43.04)</td>
</tr>
<tr>
<td>5.</td>
<td>TV (10 gm)</td>
<td>43.00 (40.98)</td>
</tr>
<tr>
<td>6.</td>
<td>TV (15 gm)</td>
<td>33.30 (35.24)</td>
</tr>
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<td></td>
<td>Combinations (An + TV)</td>
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<tr>
<td>7.</td>
<td>An + TV (2.5 gm + 2.5 gm)</td>
<td>63.50 (52.85)</td>
</tr>
<tr>
<td>8.</td>
<td>An + TV (5 gm + 5 gm)</td>
<td>60.00 (50.77)</td>
</tr>
<tr>
<td>9.</td>
<td>An + TV (2.5 gm + 7.5 gm)</td>
<td>46.60 (43.04)</td>
</tr>
<tr>
<td>10.</td>
<td>An + TV (7.5 gm + 2.5 gm)</td>
<td>53.30 (46.89)</td>
</tr>
<tr>
<td>11.</td>
<td>Control</td>
<td>96.60 (80.70)</td>
</tr>
</tbody>
</table>

CD at 5% (4.624)

Figures in parentheses indicate angular transformed value

*CD of angular transformed value.
Among three doses of *Aspergillus niger* viz. 5, 10, 15 gm/pot the least disease severity (40%) was recorded at higher level of antagonist (15 gm/pot). Likewise *Trichoderma viride* @ 15gm/pot also exhibited the least disease severity (33.3%). Among four different combinations of *Aspergillus niger* and *Trichoderma viride*, the lowest disease severity (46.6 %) was recorded where *Aspergillus niger* (2.5 gm/pot) was used in combination with *Trichoderma viride* (7.5 gm/pot). Among all the treatments whether used individually or in combination, *Trichoderma viride* (15gm/pot) proved most efficient in reducing the disease. The antagonists used individually proved more effective, than used in combination in suppression of disease. The effectiveness of both the antagonists in controlling the disease may be attributed to the suppression of pathogen by their antagonistic activity in the soil. The poor efficacy of *A. niger* and *T. viride* when used in combinations, might be due to negative antagonistic interaction between both the antagonists in the soil. The present findings are in conformity with the findings of Lacicowa and Pieta (1994) and D’Ella et al. (1998) who reported the effectiveness of various isolates of *Trichoderma* in controlling the root rot of pea caused by *Fusarium solani* f. sp. *pisi*. Similarly Wang et al. (1995) also found that *T. viride* had strong antagonistic activity towards *Fusarium solani* under *in vitro* study.

ACKNOWLEDGEMENT

Authors are grateful to the Head, Department of Plant Pathology, CCS Haryana Agricultural University, Hisar for providing the laboratory facilities. The senior author also acknowledges RAU, Bihar, Pusa, Samastipur for granting study leave during the course of investigation.

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Received for publication December 3, 2004