

In vitro Antagonistic effect of *Trichoderma* spp. against Tomato Damping off Pathogen *Pythium aphanidermatum*

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Abstract: *Pythium aphanidermatum* is an aggressive pathogen, causing damping off disease in tomato seedlings. In the present study an attempt was made to study the antagonistic effect of *Trichoderma viride* and *Trichoderma harzianum* against *P. aphanidermatum* *in vitro* condition. In dual culture method both antagonist and pathogen were grown in same potato dextrose agar (PDA) plate at 28±1°C, and the radial growth of the pathogen was compared with control (plates only with pathogen). The effect of volatile compounds released from the antagonists was studied by growing the pathogen on a PDA plate over the antagonistic fungus growing PDA plate, and the effect was determined by comparing the radial growth of pathogen in treatment and control. The bioactivity of non volatile metabolites of antagonists was detected by growing pathogen on a medium where already antagonists grew on cellophane discs for three days. In order to extract the active non volatile metabolites, antagonists were grown in potato dextrose broth for three days. Then the broth was centrifuged at 9000rpm for 20 minutes, the supernatant was passed through 0.34µm Millipore filter. Bioactivity of different dilutions of above extracts was tested by poison food technique. All the experiments were repeated three times. The results of dual culture method showed that both *T. harzianum* and *T. viride* had antagonistic effect on the growth of *P. aphanidermatum* and the effect varied significantly ($p < 0.05$), where they revealed 78.1% and 38.1% of inhibition, respectively. The volatile metabolites of both antagonists revealed inhibition on the mycelial growth at 24 hours incubation. However, amount of inhibition produced by *T. harzianum* (51.7%) was significantly ($p < 0.05$) higher than that produced by *T. viride* (37.7%) and also they lost the inhibitory effect at 48 hours. Non volatile metabolites of *T. harzianum* and *T. viride* expressed 100% and 74% inhibitions, respectively. Interestingly, in both treatments the inhibitory effect was remained even after 48 hours. Non volatile metabolic extracts of antagonists also showed growth inhibition on test pathogen and their effect varied with the concentration. *T. viride* expressed slightly higher inhibitory effect than that produced by *T. harzianum*. In conclusion, the antagonistic fungi, *T. harzianum* and *T. viride* have inhibitory effect on the plant pathogenic fungi *P. aphanidermatum in vitro*. However, further nursery bed and field studies are needed to ensure their bioactivity on *P. aphanidermatum*.