

Determination of the antifungal, biochemical and physiological characteristics of *Trichoderma* spp. isolated from onion fields of Sri Lanka

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Trichoderma species have been used as biological control agents of numerous fungal pathogens. In this study, *Trichoderma* species isolated from the soil of fifty five onion fields in different locations in the Matale and Anuradhapura districts were characterized. The antagonistic effect of three most frequently isolated *Trichoderma* spp. (Tr. 1, Tr. 3, Tr. 4) were evaluated using dual culture assays against fungal pathogens of onion *i.e.* *Fusarium* sp., *Colletotrichum gloeosporioides* and *Alternaria* sp. Four replicate plates were used for each treatment and the results analysed statistically using ANOVA. All tested *Trichoderma* spp. suppressed the mycelial growth of fungal pathogens tested. Tr. 3 caused a significantly high ($P < 0.05$) reduction of growth compared with other species showing 55.7%, 76.6% and 57.7% growth inhibition of *Fusarium* sp., *C. gloeosporioides* and *Alternaria* sp. respectively. Tr. 1 showed less inhibition amounting to 31.9 %, 53.9% and 36.9% respectively. Microscopic observations indicated that the mode of action adopted by *Trichoderma* isolates to restrict the growth of pathogens was through coiling, formation of loops and attachment of hyphal tips to the hyphae of pathogens. Tests on extracellular enzyme production by Tr. 3 and Tr. 1 using plate assays indicated the production of Chitinase and CMCase. Proteolytic activity was determined using bromocresol green dye on casein agar plates and gelatin agar plate assay. Tr. 1 and Tr. 3 produced narrow zones of hydrolysis on both casein agar plates and gelatin agar plates indicating low proteolytic activity *i.e.* 1.25 cm and 0.5 cm respectively for gelatin agar plates. Crude extracts of Tr.1, Tr.3 dissolved separately in DMSO to a final concentration of 50 mg/ml were used in well diffusion assays. After incubation, zones of inhibition of growth of *Fusarium* sp. by two *Trichoderma* isolates were observed indicating antifungal activity. The effect of volatile metabolites produced by Tr.1 and Tr.3 on *Fusarium* sp. was evaluated by placing the pathogenic fungus and each test *Trichoderma* sp on the facing halves of a petri dish and sealing with parafilm. The effect of volatile metabolites was considered as growth inhibition of *Fusarium* sp. Tr. 1 caused a 15 % and Tr. 3 a 25 % inhibition of mycelial growth of *Fusarium* sp. indicating a low level of volatile metabolite production by both species. Therefore, out of the *Trichoderma* isolates characterized, Tr.1 and Tr.3 possessed an ability to control the growth of the three onion pathogens tested and the control was achieved by both physical and biochemical activities of the test *Trichoderma* spp.

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