Management of root rot of cotton, caused by *Rhizoctonia solani*, through bioagents

G.T. Behere, V.S. Tajane, V.E. Aage and E.B. Burgoni
Department of Plant Pathology, College of Agriculture, Nagpur INDIA
Correspondence author ganeshbehere@yahoo.com

ABSTRACT

Under green house conditions, antagonistic activity of ten isolates of Trichoderma *spp.* viz., T. viride, T. harzianum, T. hamatum *and* Gliocladium virens *were tested on root rot of cotton induced by* Rhizoctonia solani. *The bioagents applied as a seed pelleting significantly reduced mortality due to* R. solani*, from 13.6 to 35.2% in cotton. The maximum percent growth inhibition was recorded from* T. viride *(GTB) 79.44%, closely followed by* T. harzianum *(GTB) 77.88% and* T. viride *(CICR) 75.22%.*

Introduction

Rhizoctonia solani Kuhn, which causes root rot and damping off in cotton, is a major problem around the world (Singh and Verma, 1988). Many of these pathogens can live in soil for long periods, particularly in presence of host debris and are accordingly difficult to control either by seed or soil application of fungicides or by cultural methods and host-plant resistance.

Among the methods of biological control available for control of soilborne diseases is the utilization of antagonistic microorganisms. Members of the genus *Trichoderma* have been widely employed as biocontrol agents (BCAs) (Wells *et. al.*, 1972) often by treating seeds prior to planting. This is a direct approach that alters the microbiological environment of the developing seed. The investigation was designed to determine the efficacy of bioagents as a seed treatment in controlling root rot of cotton induced by *R. solani*.

Experimental procedure

The pathogen causing root rot of cotton was isolated from infected root of cotton plants showing characteristic symptoms. The isolated culture were identified, purified and maintained on PDA at 27 ^{0}C. The culture was tested for its pathogenicity on cotton before it's further use. The culture of bioagents *viz.*, *T. viride* (GTB), *T. harzianum* (GTB), *Gliocladium virens* were isolated from the rhizosphere of cotton plants and other strains of *Trichoderma* spp. were obtained from Department of Plant Pathology, College of Agriculture, Nagpur.

Screening of Bioagents

Antagonistic properties of the bioagents were studied through a dual culture method. Mycelial agar plug (5 mm) were cut from the edge of one-week-old cultures of both test pathogen with a sterilized cork borer and placed 4 cm apart on PDA. Inoculated Petriplates were incubated at 27 ±1 ^{0}C in BOD chamber. Periodical observations on the growth of both the fungi were recorded for seven days. Mycelial inhibition by bioagents over control was calculated.

Seed pelleting

Effects of bioagents were studied in vivo by a seed pelleting delivery system. Bioagents was grown on PDA in Petriplates for seven days in a BOD chamber at 27 ±1 ^{0}C under continuous fluorescent light to allow profuse sporulation for five days and the spores were harvested in sterile distilled water with a camel hair brush. The concentration was adjusted to 1 x 10^8 conidia/ml. Delinted cotton seeds were immersed for five minutes in the conidial suspension (100 seeds per 500 ml) and then dried for 1-2 hours (Marshall 1982). Twenty-five seeds were sown in each pot containing 3 kg sterilized soil with three replications. Percent mortality was recorded for 30 days after sowing.

Disease Development

Sterilized sand- maize meal medium in 250 ml flask (10 gm of maize meal, 90 gm of washed sand and 15 ml of distilled water) was inoculated with two discs of 10mm diameter mycelial plug of seven days old culture grown on PDA plate. Inoculated flasks were incubated at 27 ±1 ^{0}C in a BOD chamber. After seven days of incubation, the inoculum was mixed with sterilized soil at 1:1 ratio. In each earthen pot, the inoculum mixture was applied @ 50 gm/kg of soil.

Sowing of seeds

Two days after inoculation 25 seeds pelleted with bioagents were sown in each pot, with three replications. The experiment was set up in randomized block design and percent seed germination were recorded 10 days after sowing. The seeds sown without any bioagents treatment served as check.

Disease Scoring

Based on seedlings mortality (30 days after sowing (DAS)) and reisolating the pathogen from dead seedlings on PDA, percent disease mortality and finally percent disease reduction over control were calculated. The increase in shoot and root length over other treatment and control were measured, 30 days after sowing.

Results and Discussion

All ten BCAs showed their antagonism against *R. solani in vitro*. The pattern of overgrowth by them was differed significantly between the isolates. *T. viride* (GTB) obviously showed highest degree of overgrowth (79%) closely followed by *T. harzianum*-GTB (78%) and *T. viride*-CICR (75%). The visual observations of interaction revealed that isolate *T. viride* (GTB); *T. harzianum* (GTB) and *T. viride* (CICR) started lysing the pathogen two days after contact of *R. solani*. The rest of the isolates showed intermediate overgrowth and interaction (Table 1). These observations are similar to previous reports (Elad *et. al.*, 1980, Sivan *et. al.*, 1984 and Barak *et. al.*, 1985). Similarly *G. virens* isolates were also

found effective antagonist against *R. solani* (Howell, 1982).

Seed pelleting with bioagents significantly improved seed germination. *T. viride* [GTB] increased germination by 69% followed by *T. harzianum* by 62% and *T. viride* (CICR) by 54% (Table 2). Seed treatment with BCAs for the control of *R. solani* has long been advocated although results have been variable and isolates of the same species of antagonist differ greatly in efficacy (Papavizas, 1985; Harman *et al*., 1981)

Seed pelleting with conidia of bioagents had a significant effect on disease incidence. The highest disease reduction 85% was recorded with the *T. viride* (GTB), closely followed by *T. harzianum*-GTB (79%) and *T. viride*-CICR (78%). Our studies indicate that rhizosphere isolates *T. viride* (GTB) and *T. harzianum* (GTB) have better antagonistic properties than other isolates but must be pre dominant in the rhizosphere soil in order to maintain disease free plants. Seed treatment with the BCAs had a marked effect on plant growth at 30 DAS. Treatment with *T. viride* (GTB) increased shoot length by 80% and root length by 60%, closely followed by *T. harzianum* (GTB) of shoot length 72% and root length 58% (Table 3). All treatments also exerted significant effect on growth parameters of cotton compared to control. In seed pelleting delivery system, bioagents multiplied with available moisture in root region during crop growth, thereby supporting the crop growth and reduction of root rot pathogen, *R. solani*. The results were in concordance with various workers (Elad *et. al*., 1986; Jeyarajan *et. al*., 1991; Gaikwad and Behere, 2001).

References

- Barak, R., Mazaz, A. and Chet, I. (1985). Antigenic differences among several *Trichoderma* species. *Can. J. Microbiol.* **31**: 810-816.
- Elad, Y., Kalton, A. and Chet, I. (1980). Control of *Rhizoctonia solani* in cotton by seed coating with *Trichoderma* species spores. *Pl. Soil*, **66**: 276 – 281.
- Elad, Y., Zvieli, Y. and Chet, I. (1986). Biological control of *Microphomina phaseolina* (Tassi) Goid. By *Trichoderma harzianum*. *Crop Protection*, **5**: 288-292.
- Gaikwad, S.J. and Behere, G.T. (2001). Biocontrol of wilt of safflower caused by *Fusarium oxysporium* f. sp. *carthami*. *Proceeding of Vth International Safflower Conference, Williston, N.D., U.S.A.* 2001: 63-66.
- Harman, G.E., Chet, I. and Baker, R. (1981). Factors affecting *Trichoderma harzianum* applied to seed as biocontrol agents. *Phytopathology*, **71**: 569-572.
- Howell, C.R. (1982). Effect of *Gliocladium virens* on *Pythium ultimum* and *R. solani* on damping off of Cotton seedlings. *Phytopathology,* **72**: 496-498.
- Jeyarajan, R., Ramakrishnan, G., Rajamanickam, B. and Sangita, P. (1991). Field demonstration of efficacy of *Trichoderma* as biocontrol agents for root rot disease of grains, legumes and oilseeds. *Petria,* **1**: 143.
- Lumsden, R.D. and Locke, J.C. (1989). Biological control of damping off caused by *Pythium ultimum* and *R. solani* with *Gliocladium virens* in soil less mix. *Phytopathology*, **79**: 301-305.
- Marshall, D.S. (1982). Effect of *Trichoderma harzianum* seed treatment and *R. solani* inoculum concentration on damping off of snap bean in acidic soil. *Plant Disease*, **66**: 788–789.
- Papavizas, G.C. (1985). *Trichoderma* and *Gliocladium*: Biology, ecology and potential for biocontrol. *Annu. Rev. Phytopatho,* **23**: 23-54.
- Singh, R.P. and Verma, J.P. (1988). Cotton disease problems in India. *In* Perspectives of Mycology and Plant Pathology (Eds. Agnihotri, V.P., Sarabhoy, A.K. and Kumar D.) pp. 266 – 281. *Malhotra Publishing House, Delhi*, 650 pp.
- Sivan, A., Elad, Y. and Chet, I. (1985). Biological control and effect of a new isolate of *Trichoderma harzianum* and *Pythium aphanidermatum*. *Phytopathology*, **74**: 498 – 501.
- Wells, H.D., Bell D.K. and Jaworski, O.A. (1972). Efficacy of *Trichoderma harzianum* as a biocontrol for *Sclerotium rolfsii*. *Phytopathology*, **62**: 442–447.

Table 1. In-vitro *antagonism of bio agents against* Rhizoctonia solani.

Bio agents	Mycelial growth (mm)*	Per cent inhibition of mycelial growth
T. viride (ACN)	29.20	67.55
T. viride (ACA)	30.70	55.98
T. viride (CICR)	21.40	75.22
T. viride (NRCC)	32.70	63.66
T. viride (GTB)	18.50	79.44
T. harzianum (GTB)	19.90	77.88
T. harzianum (ACA)	25.10	72.11
T. harzianum (ACN)	25.70	71.44
T. hamatum	22.90	74.55
G. virens	22.80	74.66
R. solani (Control)	99.00	----

* Average of three replications

Table 2. *Effect of seed pelleting of bio agents on the percent germination and incidence of root rot of cotton caused by* R. solani.

Treatments	Percent germination*	Percent increase over control	Percent seedling mortality*	Percent reduction over control
T. viride (ACN)	72.00	38.46	26.30	71.50
T. viride (ACA)	68.00	30.76	35.20	61.86
T. viride (CICR)	80.00	53.84	20.00	78.33
T. viride (NRCC)	68.00	30.76	35.20	61.86
T. viride (GTB)	88.00	69.23	13.60	85.26
T. harzianum (GTB)	84.00	61.53	19.00	79.41
T. harzianum (ACA)	76.00	46.15	26.30	71.50
T. harzianum (ACN)	72.00	38.46	27.70	69.98
T. hamatum	76.00	46.15	26.30	71.50
G. virens	76.00	46.15	26.30	71.50
R. solani (Control)	52.00	----	92.30	-----
SE.d	0.62	-----	1.13	-----
CD at 5%	1.30	-----	2.36	-----

* Average of three replications and 5 seedlings (Percent germination were taken 10 days after sowing)

Table 3. *Effect of seed pelleting with bio agents on plant biometrics at 30 DAS.*

Treatments	Shoot length (cm)*	Percent increase over control	Root length (cm)*	Percent increase over control
T. viride (ACN)	17.43	24.50	12.30	23.49
T. viride (ACA)	17.23	23.07	11.53	15.76
T. viride (CICR)	22.66	61.85	14.10	41.56
T. viride (NRCC)	16.76	19.71	12.00	20.48
T. viride (GTB)	25.13	79.50	15.90	59.63
T. harzianum (GTB)	24.13	72.35	15.76	58.23
T. harzianum (ACA)	18.86	34.71	12.50	25.50
T. harzianum (ACN)	17.96	28.28	12.66	27.10
T. hamatum	22.23	58.78	14.10	41.56
G. virens	21.96	56.85	14.00	40.56
R. solani (Control)	14.00	-----	09.96	----
SE.d	0.23	-----	0.29	-----
CD at 5%	0.49	-----	0.61	-----

* Average of three replications and 5 seedlings.