

Table 3. Reaction of sodic soil tolerant rice cultures against leaffolder and yellow stem borer (Mean of two season experiments)

Rice culture/ variety	Leaffolder damage rating	Stem borer white ear damage rating
IRRI 2028	7.0 a	7.0 a
IRRI 2030	7.0 a	5.6 b
IRRI 2011	3.6 c	5.0 b
IRRI 2122	3.0 c	3.0 c
PTB 33	1.0 d	1.6 d
CO 43	5.0 b	5.6 b

Mean of 5 replications

In a column, means followed by the same letter are not significantly different by DMRT (P=0.05)

moderately resistant culture IRRI 2122 (61.0) and IRRI 2028 (86.0). The average nymphal duration on moderately resistant sodic soil cultures was longer than other cultures indicating some antibiotic effect. In the sodic soil tolerant moderately resistant variety, the nymphal duration was delayed IRRI 2028 (16.20 days), IRRI 2122 (16.0) when compared with CO 43 (13.0 days). The

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BIOLOGICAL CONTROL OF STEM ROT OF TOMATO CAUSED BY *Sclerotium rolfsii* Sacc

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ABSTRACT

Nine antagonists were screened for the antagonistic effect against tomato stem rot pathogen. *In vitro* studies showed that the *Bacillus subtilis* and *Pseudomonas fluorescens* were highly significant in inhibiting the mycelial growth and sclerotial production of the pathogen. Next to these two antagonists, *Trichoderma harzianum* and *T. viride* were effective in reducing the sclerotial size and germination. Pot culture studies indicated that the soil amendment with *T. harzianum* resulted in better plant stand upto 80 per cent at 60 DAS followed by *T. viride* and *P. fluorescens*.

KEY WORDS : Tomato, Stem rot, *S. rolfsii*, Biological control

The stem rot disease of tomato caused by *Sclerotium rolfsii* Sacc. is distributed in tropical and subtropical regions of the world and is common in India, Southern United States, Central America, Africa, Australia and the countries surrounding the mediterranean. In addition to tomato, the pathogen causes severe damage to green bean, lima bean, onion, pepper, potato, watermelon, Southern Pea and Sweet potato

fecundity was significantly higher on susceptible cultures CO43 (203 no.) IRRI 2011 (163); IRRI 2030 (160) as against IRRI 2028 (137). The female adult longevity was also higher in CO43 (12.40 days) as compared to shorter period in IRRI 2122 (6.0 days) (Table.2). All the sodic soil cultures was susceptible to LF and YSB except IRRI 2122. The culture IRRI 2028 was found to be highly susceptible to both pests by recording a grade of 7.0 as against 3.0 in IRRI 2122 (Table 3).

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(Aycock, 1966). The management strategies using chemicals have been reported so far. But the chemicals pose environmental problems in addition to escalating cost. Hence, an experiment was carried out to manage the disease with biocontrol agents since these agents are cheap and also very effective for the management of soil borne diseases (Cook and Baker, 1983).

MATERIALS AND METHODS

The pathogen *S. rolfii* was isolated from collar region of diseased plants by tissue segment method (Rangaswami, 1972) and purified by hyphal tip method and maintained on potato dextrose agar slants.

In vitro screening of antagonists against *S. rolfii*

The antagonistic fungal species *T. viride*, *T. harzianum*, *T. hamatum*, *T. koningii*, *T. pseudokoningii*, *T. longibrachiatum* and *Gliocladium virens* and two bacterial antagonists *B. subtilis* and *P. fluorescens* were used to test their antagonism against *S. rolfii* by dual plate technique (Dennis and Webster, 1971).

A 9 mm disc of the antagonistic fungus was cut out from the 7 days old culture with the help of sterile cork borer and placed at one end of the Petri dish containing 15 ml potato dextrose agar medium. In the opposite end a similar disc of pathogen *S. rolfii* was placed. The radial growth of pathogen and antagonistic fungus was measured after 96 hours of incubation.

In case of bacterial antagonist, it was streaked for 1 cm length instead of placing a culture disc. The plates were incubated at room temperature for 96 hours. Three replications were maintained for each treatment. The growth of the pathogen and antagonistic organisms were recorded.

The per cent inhibition of growth was calculated by the formula of Vincent (1927) as follows.

$$I = \frac{100(C-T)}{C}$$

- I = Inhibition per cent
 C = Germination / Growth in control
 T = Germination / Growth in treatment

Effect of antagonists against sclerotial production, size and germination

The pathogen and the antagonistic fungi / bacteria were grown in dual culture on potato dextrose agar medium. After 240 hours of incubation the mature sclerotia were harvested from each plate with the sterile forceps and counted. The size was measured with a use of calibrated ocular micrometer at 10 x X 15 x magnification in a compound microscope.

The mature sclerotia picked up from each treatment were placed over potato dextrose agar medium at the rate of ten sclerotia / plate with equal space. The plates were incubated for 24 hours and examined under a stereo microscope to record the sclerotial germination and production of hyphae.

Evaluation of biocontrol agents against *S. rolfii* by soil application under pot culture conditions.

Pots of 30 x 20 cm size were filled with soil and *T. Viride*, *T. harzianum*, *T. hamatum*, *T. koningii*, *T. pseudokoningii*, *T. longibrachiatum* and *G. virens* multiplied in sand maize medium were added

Table 1. Efficacy of antagonistic organisms against *S. rolfii* (dual plate technique)

Treatments	* Linear growth of antagonists / pathogen mm	% inhibition over control
<i>B. subtilis</i>	12.7 (3.55)g	85.89
<i>P. fluorescens</i>	13.7 (3.69)h	84.78
<i>T. viride</i>	23.0 (4.79)e	74.44
<i>T. harzianum</i>	23.3 (4.83)e	74.11
<i>T. koningii</i>	24.7 (4.96)d	72.56
<i>T. longibrachiatum</i>	37.0 (6.08)e	58.89
<i>T. pseudokoningii</i>	37.7 (6.13)c	58.11
<i>T. hamatum</i>	42.0 (6.48)b	53.33
<i>G. virens</i>	38.7 (6.21)c	57.00
Control	90.0 (9.48)a	0.00

* Mean of 3 replications

Data in parentheses indicate root transformed values

In a column, mean followed by a common letter are not significantly different at 5% level by DMRT.

at the rate of 5g/kg of soil. Talc based formulation of *P. fluorescens* and peat based formulation of *B. subtilis* was added at the rate of 0.5 g/kg of soil. The antagonists were applied to the soil and allowed for their multiplication in soil. One week after, the pathogen *S. rolfii* multiplied in sand maize medium was added at the rate of 50 g / kg of soil to each pot. The tomato seedlings were maintained at the rate of 4 per pot. Seedlings in pot with *S. rolfii* inoculum alone served as control. Three replications were maintained for each treatment. The experiment was set up in a completely randomised block design. The percent mortality was recorded at 30 days, 45 days and 60 days after sowing.

RESULTS AND DISCUSSION

SCREENING OF ANTAGONISTS AGAINST *S. rolfii* UNDER IN VITRO CONDITION

Inhibition of mycelial growth

All the nine antagonists tested inhibited mycelial growth of the pathogen by more than 50 per cent. Maximum inhibition of 85.8 per cent and 84.7 per cent were recorded by bacterial antagonists *B. subtilis* and *P. fluorescens* (Table 1), followed by fungal antagonists *T. harzianum* and *T. viride*. The antagonistic effect of *B. subtilis* against *S. rolfii* growth has been reported by Narain and Mohanty (1983). Kwee and Keng, (1990) suggested that multiple mechanisms involving mycoparasitism, antibiosis, lysis and hyphal interference may be implicated in the interaction between *T. harzianum* and *S. rolfii*

Effect of sclerotial production, size and germination

Of the nine antagonists tested, the bacterial antagonists *B. subtilis* and *P. fluorescens* completely inhibited the sclerotial production. Among the *Trichoderma* spp. antagonism of *T. harzianum* recorded lowest number of sclerotia (11.7 per cent) followed by *T. viride* and *G. virens* (Table 2). The sclerotial size and germination was also reduced significantly by *T. harzianum*. Inhibition of sclerotial production by the bacterial

antagonists led to lower inoculum potential thereby resulted in lesser disease incidence. The ability of *T. harzianum* to reduce the mycelial growth, sclerotial formation and sclerotial germination has been reported by D'Ambra and Ferrata (1984).

Evaluation of biocontrol agents and against *S. rolfii* by soil application under pot culture condition.

Among the antagonists, *T. harzianum* was

Table 2. Effect of antagonistic organisms on sclerotial production, sclerotial size and germination.

Treatments	* Number of Sclerotia per plate	* Size of Sclerotia (m)	* Germination of Sclerotia (%)
<i>B. subtilis</i>	0.0 (0.0) ^a	0.0 (0.0) ^a	0.0 (0.0) ^a
<i>P. fluorescens</i>	0.0 (0.0) ^a	0.0 (0.0) ^a	0.0 (0.0) ^a
<i>T. viride</i>	24.6 (4.9) ^a	1067.3 (32.7) ^a	76.7 (61.2) ^a
<i>T. harzianum</i>	11.7 (3.4) ^b	1271.7 (35.7) ^a	56.7 (48.8)
<i>T. koningii</i>	31.3 (5.6) ^a	1222.3 (34.9) ^a	76.7 (61.2) ^a
<i>T. longibrachiatum</i>	28.0 (5.3) ^a	1228.3 (35.0) ^a	70.0 (56.8) ^a
<i>T. pseudokoningii</i>	42.7 (6.5) ^a	1151.7 (33.9) ^a	76.7 (61.2) ^a
<i>T. hamatum</i>	27.7 (5.3) ^a	1206.7 (34.7) ^a	80.0 (63.4) ^a
<i>G. virens</i>	24.3 (4.9) ^a	1203.3 (34.6) ^a	90.0 (71.6) ^a
Control	61.3 (7.8) ^a	1230.0 (35.1) ^a	90.0 (71.6) ^a

* Mean of 3 replications

Data in parentheses indicate square root and arcsine transformed values

In a column, means followed by a common letter are not significantly different at 5% level by DMRT

Table 3. Effect of soil application of biocontrol agents against *S. rolfii* (pot culture studies)

Treatments	Stem rot incidence (%)		
	30 DAS	45 DAS	60 DAS
<i>B. subtilis</i>	26.7 ^{de} (31.1)	33.3 ^d (35.2)	33.3 ^d (35.2)
<i>P. fluorescens</i>	20.0 ^{cd} (26.6)	26.7 ^c (31.1)	26.7 ^c (31.1)
<i>T. viride</i>	20.0 ^{cd} (26.6)	26.7 ^c (31.1)	26.7 ^c (31.1)
<i>T. harzianum</i>	6.7 ^{bc} (15.0)	20.0 ^b (26.6)	20.0 ^b (26.6)
<i>T. koningii</i>	26.7 ^{de} (31.1)	33.3 ^d (35.2)	40.0 ^e (39.2)
<i>T. longibrachiatum</i>	46.7 ^e (43.1)	46.7 ^e (43.1)	46.7 ^e (43.1)
<i>T. pseudokoningii</i>	26.7 ^{de} (31.1)	33.3 ^d (35.2)	33.3 ^d (35.2)
<i>T. hamatum</i>	40.0 ^{de} (39.2)	46.7 ^e (43.1)	53.3 ^f (46.9)
<i>T. virens</i>	40.0 ^{de} (39.2)	46.7 ^e (43.1)	46.7 ^e (43.1)
Control (uninfested)	0.0 ^a (1.8)	6.7 ^a (15.0)	6.7 ^a (15.0)
Control (infested)	40.0 ^{de} (39.2)	46.7 ^e (43.1)	53.3 ^f (46.9)

¹ Mean of 3 replications

Data in parentheses indicate arcsine transformed values

In a column, mean followed by a common letter are not significantly different at 5% level by DMRT.

highly effective against the stem rot pathogen *S. rolfii* thereby recording least stem rot incidence of 20.0 per cent at 60 DAS (Table 3). The next best effective treatment were *T. viride* and *P. fluorescens* which recorded 26.7 per cent and 26.7 per cent incidence respectively. This indicated that the antagonists when applied to the soil along with appropriate food base were able to survive for long time and gave protection against the pathogen,

even upto 60 DAS. Soil inoculation of *T. viride* and *T. harzianum* multiplied in FYM and applied to the soil at the rate of 5g/kg of soil at the time of inoculation of pathogen was reported to give significant reduction in incidence of *S. rolfii* in groundnut (Muthamilan, 1989). The results from the present study indicated that both *T. harzianum* and *P. fluorescens* survive well in rhizosphere of tomato which may be due to its improved capacity to compete with the root invaders (Scher *et al.*, 1988).

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