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# INTEGRATED MANAGEMENT OF COLLAR ROT OF TOMATO

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**ABSTRACT**

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An integrated experiment was carried out with bio-agents, fungicide, plant growth hormone (Vegemax), soil amendment to find out a suitable strategy for the management of collar rot caused by *Sclerotium rolfsii* of tomato under inoculated condition at BINA farm, Mymensingh during the summer season of 2008-09. Apparently healthy seeds of Binatomato-5 treated with hot water were used for the experiment. All treatments showed positive effect in controlling the disease. Integrated control measure such as Agrizeb + Mustard oil cake + *T. harzianum* gave the highest reduction in disease index (76.7%) and severity (44.4%) of collar rot. All treatments increased plant height and fruit yield compare to control except the treatment Tilt + Mustard oil cake + *Trichoderma harzianum*. This treatment reduced plant growth as well as yield of tomato. So, the integrated treatment Agrizeb + Mustard oil cake + *T. harzianum* was very effective and eco-friendly manner in controlling the disease. Moreover, this restored soil fertility and increased yield. But incase of fungicides has no such advantages except disease control. The integrated treatment Agrizeb + Mustard oil cake + *T. harzianum* play an important role in improving soil fertility rendering the crop more tolerant to disease and thus causes the high yielding ability of tomato.

***Key words:*** *collar rot, integrated management and tomato*

## INTRODUCTION

Collar rot occurs world wide but is most important in tropical and sub tropical areas. The pathogen *S. rolfsii* sacc attacks more than 500 species of plant including many economically important vegetables, ornamental and field crop (Anahosur 2001). The disease has several other common names including southern stem rot, southern wilt and southern blight. *S. rolfsii* appears every year in all tomato growing districts of Bangladesh. As all required optimum climatic conditions are prevailing in this zone and further the fungus being poly phages, hence the disease occurs every year. Plants of any age may be attacked. Under favorable conditions of the disease may out break in epiphytotic forms and can either affects its yield and quality or cause total failure of the crop incurring substantial losses to the economy of the country. *S. rolfsii* is chiefly soil borne pathogen and prefers damp, especially water logged conditions (Fakir 2000). As the summer season of Bangladesh is wet and eventually caused water logging in low laying areas, so the incidence of collar rot has been found more in this season (Babar 1999). The pathogen likes soft tissues and causes rot of tissues adjacent to soil level termed collar zone causing death by disrupting translocation of food from the top to the root zone. That is why, control of the disease is very essential. Bio-agent is very effective to control the pathogen and restore the soil fertility without disturbing the components of the nature. On the other hand, chemical control is a very important method for complete eradication of the pathogen and in case of severe attack adequate dose of chemicals is very useful and less hazardous for the nature. Under this concept the present study has been under taken to evaluate the effectiveness of fungicides, soil amendments and bio-agents against the disease.

## MATERIALS AND METHODS

The experiments were carried out in the research laboratory and the experimental field of Bangladesh Institute of Nuclear Agriculture (BINA), Mymensingh. The experimental area falls under the subtropical climate, characterized by heavy rainfall during April to September and scanty of rainfall during October to March Khan *et al.* (1998). Tomato seeds and/or 4-5 weeks old seedlings of the variety Binatomato-5 were used throughout the investigation. Thirty day old seedlings were inoculated by soil drenching method with 7 day old culture of the pathogen.

### *Computation of disease index*

The disease index was computed by adopting (1-9) scale for *S. rolfsii* (ICRISAT (Nene and Thapliyal, 1982).

| Scale | Mortality | Reaction |
|---|---|---|
| 1 | 0 | Resistant (R) |
| 2-3 | 10 or less | Moderately Resistant (MR) |
| 4-5 | 11-20 | Tolerant (T) |
| 6-7 | 21-50 | Moderately Susceptible (MS) |
| 8-9 | 51-above | Susceptible (S) |

The disease incidence was evaluated using the following formulae:

$$\text{Percent disease index (PDI)} = \frac{(\text{Sum of total scores}) \times 100}{(\text{Maximum grade}) \times \text{Total number of plants assessed}}$$

***Field screening of mutants/varieties for disease resistance against collar rot of tomato***

The seeds of fourteen mutant lines/varieties were collected from BINA, Mymensingh, and Bangladesh Agricultural Research Institute (BARI), Joydebpur, Gazipur and Seed store, Mymensingh. The seeds were stored in refrigerator at 5-7$^0$C until use for subsequent studies.

Screening of tomato mutant lines/varieties for disease resistance was done through induced disease development by inoculating the plants with *S. rolfsii*. The experiment was carried out in the farmers field of Mymensingh, Magura and Ishurdi following two factors randomized complete block design with three replications during the summer season of 2009. Thirty day old healthy seedlings grown with intensive care in aluminum trays were transplanted in the field in the afternoon followed by watering. Sixteen healthy seedlings of each mutant/variety were transplanted in each plot with a spacing of 50 cm between rows and plants. All seedlings of one month old plants/plot were inoculated by collar region inoculation method with 7 day old culture of *S. rolfsii*. After inoculation, the collar region of the seedlings were covered with moist cotton. Disease severity of plants was recorded at 60 days after inoculation.

***In-vitro evaluation of fungicides***

Six concentrations *viz.* 125, 250, 500, 750, 1000 and 1500 ppm of seven fungicides such as Bavistin, Tilt, Dithane M-45, Securre, Agrizeb, Timsen and Haymoxy were tested to observe the percentage inhibition of mycelial growth of *S. rolfsii* by using poisoned food technique (Nene and Thapliyal, 1979). Required amount of fungicides of each concentration was thoroughly mixed with melted PDA. Thus, 100 ml poisoned PDA for each concentration was prepared (50 ml double concentrated PDA + 50 ml fungicidal solution). Plates of 9 cm diameter each containing 20 ml medium were maintained for each of the concentrations ranging from 125-1500 ppm. For each control plate PDA was prepared by mixing distilled water and maintained against each of the fungicide.

***In-vitro screening of antagonists***

Five antagonists *viz. T. harzianum* ($PT_{02}$), *T. harzianum* (BPT $_{03}$), *T. harzianum* (BPT $_{09}$), *T. viride* ($BPT_{12}$) and *T. harzianm* (BPT $_{17}$) were screened against the pathogen.

***Growth inhibition technique***

A mycelial block of the pathogen and the antagonist (same size and age) were placed in an inverted position at 5 cm apart on PDA medium. Linear growth (mm) of the pathogen along with control against all antagonists were measured. Mycelial growth of the pathogen at 24 hours interval up to 7 days was recorded. The growth inhibition in percentage was calculated by using the following formulae of Asharafuzzaman and Hossain (1992).

$$I = \frac{C - t}{C} \times 100$$

Where, C = average growth (mm) in the control, t = average growth (mm) in the treatment,
I = Percentage of growth inhibition

***Integrated management of collar rot of tomato***

An integrated experiment was carried out with bio-agents and clean seed treated with hot water, fungicides, vegemax and soil amendment to find out a suitable strategy for the management of collar rot of tomato at BINA farm, Mymensingh and BINA sub-station farms, Magura and Ishurdi during the summer season of 2008-09. Apparently healthy seeds of Binatomato-5 was treated with hot water.

The treatments were as follows:

$T_1$ = Agrizeb + Mustard Oil Cake (MOC)
$T_2$ = *T. harzianum* + MOC + Vegemax
$T_3$ = MOC + *T. harzianum*
$T_4$ = Agrizeb + MOC + *T. Harzianum*
$T_5$ = MOC + Agrizeb + Vegemax
$T_6$ = Tilt + *T. harzianum* + MOC
$T_7$ = Control (Sterilized water)

***Details of the treatments***

Mustard Oil cake (150kg/ha) paste made by soaking it for 3 days in water were mixed in the upper 15 cm of rhizosphere soil of the tomato seedling 10 days before planting. Ten days after transplanting *T. harzianum* 20g/plant) as oat culture were thoroughly mixed with soil around the plants.

The fungicidal solutions were prepared in water, Agrizeb 80 WP (3g/L) and Tilt (0.5ml/L) were applied twice at @ 200 ml plant$^{-1}$ 24 hours before transplanting and 24 hours before inoculation. The chemicals were applied to the rhizosphere of tomato plants as fungicidal solution using a beaker by soil drenching method. Data on the incidence and severity of collar rot were recorded at 15, 30 and 45 days after inoculation by visual observation of symptoms. Number of infected plants plot$^{-1}$, disease severity, plant height and yield plot$^{-1}$ were recorded.

## RESULTS

### *Field screening of mutants/varieties of tomato against collar rot*

The response of eleven mutants/varieties tested against collar rot of tomato under inoculated conditions at Mymensingh, Ishurdi and Magura is presented in Table 1. The percent disease index (PDI) and disease severity (DS) differed significantly among the tested materials at all the locations. At Mymensingh PDI and DS of the disease ranged from 33.3-70.8% and 6.0-8.3, respectively. The percentage of wilted plants varied from 33.3-54.2% in Binatomato-5, Binatomato-3, BARItomato-5, TM-105, TM-133, TM-219, TM-131 and TM-134 and they were graded as moderately susceptible while the mutant/varieties CLN-2026, BARItomato-4 and Binatomato-2 were identified as susceptible. However, the highest percentages of wilted plants were found in the mutant CLN-2026 to collar rot.

At Ishurdi, only the variety Binatomato-5 was graded as tolerant. The highest percent disease index (100%) and disease severity (9.0) was recorded in the mutant line TM-105 which was statistically similar to TM-131, TM-133, CLN-2026, Binatomato-2 and Binatomato-3 while the lowest PDI and DS were recorded in the variety Binatomato-5. The second lowest disease index was recorded in the mutant TM-134 while the second highest disease index was recorded in the mutant TM-133. At Magura, considerably the highest PDI and DS were recorded in the mutant CLN-2026 followed by TM-134 while the lowest PDI and DS were observed in the variety Binatomato-5 followed by TM-110, TM-131, Binatomato-2, Binatomato-3 and BARItomato-5. Binatomato-5, BARItomato-4, Binatomato-3 and Binatomato-2 showed tolerant reaction to the disease at Magura.

Table 1. Disease index and severity of some selected mutants/varieties of summer tomato to collar rot at Mymensingh, Ishurdi and Magura

| Mutants/variety | Mymensingh | | | Ishurdi | | | Magura | | |
|---|---|---|---|---|---|---|---|---|---|
| | PDI | DS | DR | PDI | DS | DR | PDI | DS | DR |
| Binatomato-5 | 33.3 | 6.0 | MS | 44.42e | 4.6e | T | 20.0d | 5.0ef | T |
| BARItomato-4 | 58.3 | 7.6 | S | 79.9bcd | 9.0c | S | 31.1bc | 6.3def | T |
| BARItomato-5 | 54.2 | 7.0 | MS | 77.7cd | 8.0b | S | 24.4bcd | 5.3bc | MS |
| Binatomato-3 | 56.3 | 7.0 | MS | 95.5ab | 9.0a | S | 24.4bcd | 5.0ef | T |
| Binatomato-2 | 64.6 | 8.3 | S | 86.6abcd | 9.0a | S | 24.4bcd | 5.4def | T |
| TM-133 | 43.8 | 6.6 | MS | 97.8a | 9.0a | S | 31.8bc | 6.3bc | MS |
| CLN-2026 | 70.8 | 8.3 | S | 93.3abc | 9.0a | S | 48.9a | 7.6a | S |
| TM-219 | 37.6 | 6.3 | MS | 73.3d | 8.6a | S | 35.6b | 7.0ab | MS |
| TM-131 | 41.6 | 7.0 | MS | 86.6abcd | 9.0a | S | 28.9bcd | 6.0cde | MS |
| TM-105 | 45.8 | 7.0 | MS | 100.0a | 9.0a | S | 35.6b | 6.0cde | MS |
| TM-134 | 45.8 | 6.6 | MS | 45.42- | - | - | 46.7a | 7.0ab | MS |
| Level of sign. | 0.01 | 0.01 | | 0.01 | 0.01 | | 0.01 | 0.01 | |

T = Tolerant, MS = Moderately Susceptible, S = Susceptible

The percent disease index and severity of collar rot of the mutants/varieties at different locations differed significantly. However the lowest percent disease index (31.54%) and severity (7.1) was recorded at Magura while the highest percent disease index (80.05%) and severity (8.4) was recorded at Ishurdi. Mymensingh ranked second in position. The mutants/varieties showed susceptible reaction to the disease at all the locations

Table 2. Mean disease index and severity of some mutants/varieties of summer tomato to collar rot at different locations

| Locations | Disease index (%) | Disease severity | Disease reaction |
|---|---|---|---|
| Mymensingh | 50.63b | 7.8b | S |
| Ishurdi | 80.05a | 8.4a | S |
| Magura | 31.54c | 7.2c | S |
| LSD(0.01) | 12.86 | 0.52 | |

S = Susceptible

### *Evaluation of fungicides*

All the fungicides used during the present studies showed a gradual decline in growth of *S. rolfsii* with increasing in concentration of medium except Agrizeb, Tilt and Haymoxy (Fig. 1). Agrizeb and Tilt @ 125 ppm were found to be the most effective fungicides and gave 100% reduction in myceal growth of *S. rolfsii.* Dithane M-45 was the next effective fungicide however, its efficacy was not significantly different from that of Agrizeb and Tilt. It gave >70% reduction in 125ppm. Secure 125 ppm produced >50% reduction in growth. Carbendazim produced >50% reduction in growth when used @ 1000 ppm. Timsen 1000 ppm produced only

>5% reduction in growth of *S. rolfsii*. Haymoxy was found to be not effective in reducing the mycelial growth of *S. rolfsii* at the lower and higher concentrations.

***Screening of fungal antagonists***

All the five isolates of fungal antagonists caused significant inhibition in diametrical growth of the test pathogen *S. rolfsii*. The magnitude of inhibition varied according to the species (Table 3**)**. Among them, *T. harzianum* ($BPT_{17}$) was the most effective antagonist followed by *T. viride* ($BPT_{12}$) in reducing the radial growth of the pathogen. However, *T. harzianum* ($BPT_{17}$) and *T. viride* ($BPT_{12}$) were on par with each other in respect to radial growth and significantly superior to all other isolates and also of control. *T. harzianum* ($BPT_{02}$) was the next best. In addition, *T. harzianum* ($BPT_{03}$) and *T.* harzianum ($BPT_{09}$) also showed significant reduction in radial growth compared to control. *T.* harzianum ($BPT_{09}$) was the least effective having 90.18% inhibition in radial growth over control. A reduction in the growth of *S.rolfsii* was noticed when it was placed with two antagonists (*T. harzianum* and *T. viride*) 5 cm apart on PDA medium in separate petridishes within 4 days of inoculation, both the antagonists *T. harzianum* and *T. viride* overgrew *S. rolfsii* colony (Plate 1 & 2).

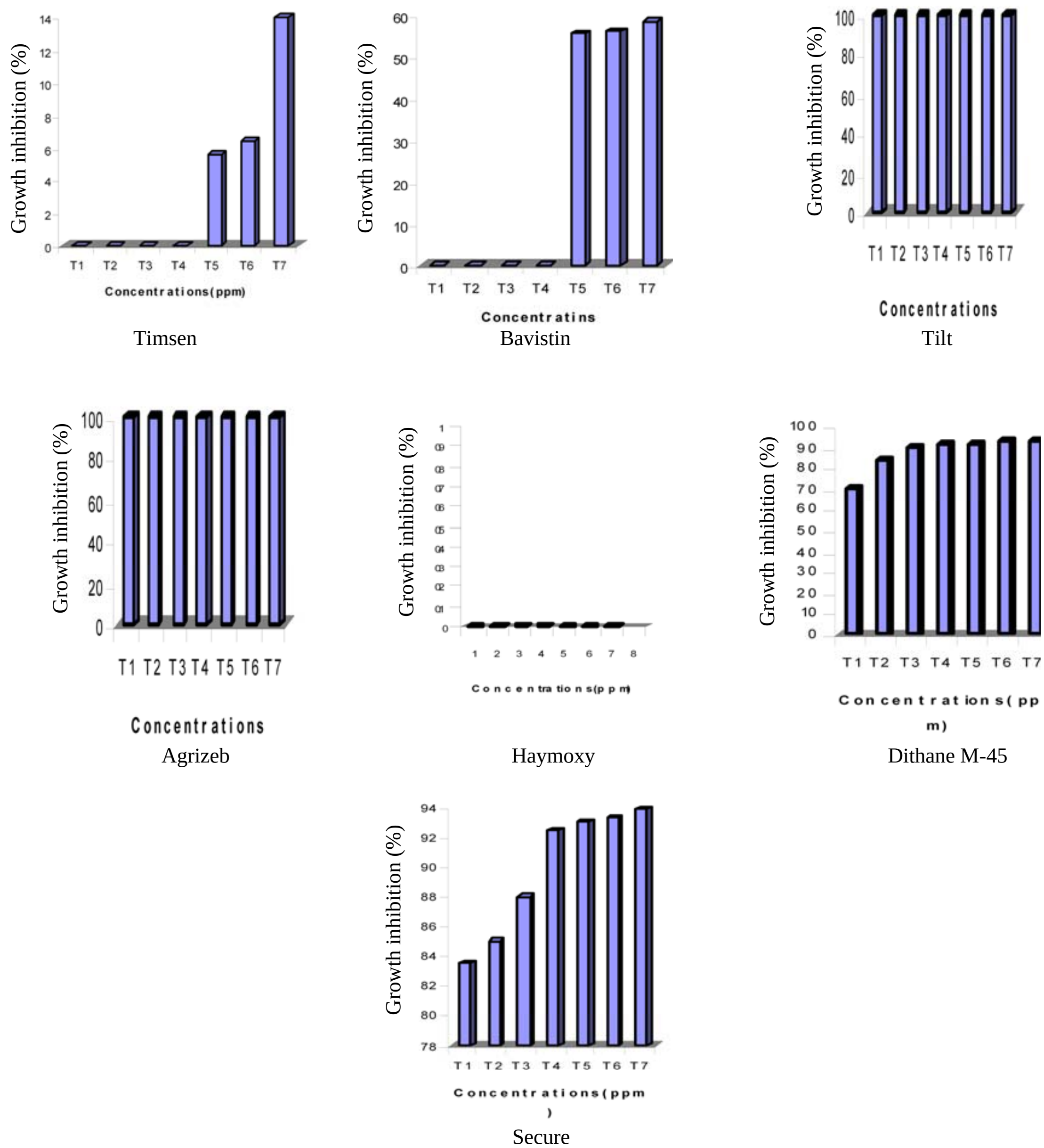


Fig. 3. Percent growth inhibition of *S. rolfsii* on PDA containing different concentrations of fungicides
$T_1$ = 125, $T_2$ = 250, $T_3$ = 500, $T_4$ = 750 ppm
$T_5$ = 1000 $T_6$ = 1250 and $T_7$ = 1500 ppm

Table 3. Effect of preliminary screening of fungal antagonists

| Antagonists | *S. rolfsii* | |
|---|---|---|
| | Linear growth (mm) | Inhibition (%) of radial growth over control |
| *T. harzianum* ($BPT_{02}$) | 7.43cd | 91.74 |
| *T. harzianum* ($BPT_{03}$) | 7.73c | 91.85 |
| *T. harzianum* ($BPT_{09}$) | 8.83b | 90.18 |
| *T. viride* ($BPT_{12}$) | 7.00d | 92.22 |
| *T. harzianum* ($BPT_{17}$) | 6.17e | 93.14 |
| Control | 90.00a | --- |
| Level of significance | 0.01 | |

***Integrated management of collar rot of tomato***

The result of percent disease index and severity of Binatomato-5 at 30 and 60 days after inoculation (DAI) are presented in table 4. In general all the treatments had substantial positive effect on the reduction of percent disease index over control at both the dates of data collection. Consequently the lowest disease index and severity were recorded in plants receiving $T_4$ treatment (Index = 19.4%, severity = 5.0) followed by $T_1$ treatment (Index = 20-24.65%, severity = 5.0,6.0) at both the dates of observations. The third lowest percent disease index was observed in $T_6$ treatment (Tilt + *T. harzianum* + MOC = 33.0%). The highest percent disease index was recorded in untreated control (inoculated) treatment during both the dates of data recording. The percent disease index did not flare up from 30 DAI to 60 DAI at $T_4$ treatment.

Significant variation among the treatments was observed on the reduction of percent disease index and disease severity **(**Fig 1and 2**).** The highest reduction was observed in $T_4$ treatment. $T_2$ (*T. harzianum* + MOC + Vegemax) and $T_3$ (MOC + *T. harzianum* + HWT) treatments showed statistically similar effect. However, $T_3$ was more effective than $T_2$ while $T_6$ was the more effective than $T_5$ treatment in reducing the percent disease index and severity.

Table 4. Integrated control of *Sclerotium rofsii* infection of tomato

| Treatments | 30 Days after inoculation | | | 60 Days after inoculation | | |
|---|---|---|---|---|---|---|
| | PDI | DS | DR | PDI | DS | DR |
| $T_1$ = Agrizeb + MOC | 20.8ef | 5.0 | T | 24.6.0e | 6.0 | MS |
| $T_2$ = *T. Harzianum* + MOC + Vegemax | 41.7b | 6.2 | MS | 47.9b | 7.0 | MS |
| $T_3$ = *T. harzianum* + MOC | 33.3c | 6.0 | MS | 44.4b | 6.5 | MS |
| $T_4$ = Agrizeb **+** MOC + *T. harzianum* | 19.4f | 5.0 | T | 19.4f | 5.0 | T |
| $T_5$ = Agrizeb + MOC + Vegemax | 27.8d | 6.0 | MS | 38.5c | 6.3 | MS |
| $T_6$ = *T. harzianum* + MOC + Tilt | 25.0de | 6.0 | MS | 33.0d | 6.0 | MS |
| $T_7$ = Control (Sterilized water) | 74.9a | 9.0 | S | 83.3a | 9.0 | S |
| Level of significance | 0.01 | 0.01 | | 0.01 | 0.01 | |

In a column, figures bearing common letter(s) do not differ significantly, PDI = Percent disease index, DS = Disease severity, MOC = Mustard oil cake

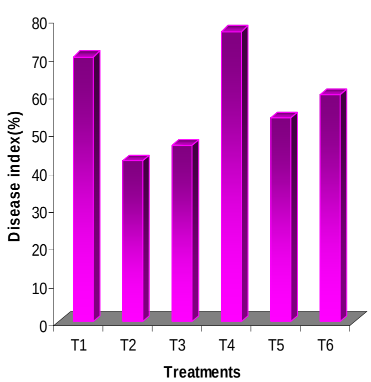


Fig.1. Effect of treatments on reduction of percent disease index at 60 days after inoculation

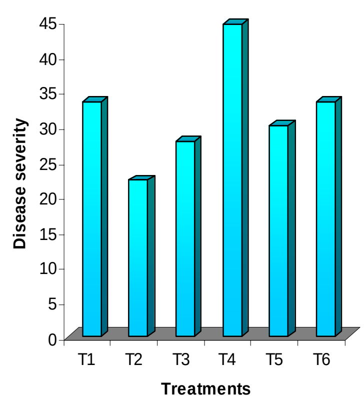


Fig.2. Effect of treatments on reduction of percent disease severity at 60 days after inoculation

$T_1$ = Agrizeb + MOC + HWT
$T_2$ = *T.h* + MOC + Vegemax
$T_3$ = MOC + *T.h* + HWT
$T_4$ = Agrizeb **+** MOC **+** HWT + *T.h*
$T_5$ = MOC + Agrizeb + Vegemax
$T_6$ = Tilt + *T.h* + MOC + HWT
*T.h* = *Trichoderma harzianum*
MOC = Mustard Oil Cake

All treatments increased plant height and fruit yield per plot compared to control except $T_6$ treatment in all the locations (Table 5). Though the highest plant height was observed in $T_2$ treated plot while the highest fruit yield was recorded in $T_4$ treated plot where Agrizeb, Mustard oil cake and *T. harzianum* were applied in the soil. $T_6$ treatment reduced the plant height and showed no fruit development though the fungicide (Tilt) resulted in no mycelia growth of *S. rolfsii* in PDA medium in the petri plates.

Table 5. Effect of treatment combination on plant height and yield against collar rot of tomato at Mymensingh

| Treatments | Percent shoot length and yield increased over control | |
|---|---|---|
| | Plant height | Yield |
| $T_1$ = Agrizeb + Mustard oil cake | 49.6 | 19.7 |
| $T_2$ = *T. harzianum* + Vegemax + Mustard oil cake | 57.8 | 22.2 |
| $T_3$ = Mustard oil cake + *T. h*arzianum | 49.5 | 19.4 |
| $T_4$ = Agrizeb **+** *T. harzianum* + Mustard oil cake | 51.8 | 37.0 |
| $T_5$ = Agrizeb + Vegemax + Mustard oil cake | 53.5 | 22.0 |
| $T_6$ = Tilt + *T. h*arzianum + Mustard oil cake | -20.3 | -- |

**DISCUSSION**

The findings of the present study revealed that the tested materials showed different types of reaction of disease under artificially inoculated conditions. The variety Binatomato-5 was resistant to collar rot at Magura and Ishurdi while in Mymensingh it showed moderately susceptible reaction. In the present study variation in the prevalence of disease index and severity recorded is in consistence with the observation of other workers (Hossain 1991; Meah *et al*. 1998). Host resistance is a type of biological control in which host itself plays the role of an antagonist. The view is supported by the report of Singh (2002) who stated that resistant variety can be the most simple, practical, effective and economical method of plant disease management.

In vitro screening experiments indicated that all the biocontrol agents *viz. T. harzianum* ($BPT_{02}$), *Trichoderma* sp ($BPT_{03}$), *T. harzianum* ($BPT_{09}$), *T. viride.* ($BPT_{12}$) *T. harzianum* ($BPT_{17}$). Were found effective in inhibiting the growth of *S. rolfsii*. The findings was in conformity with that of Begum *et al.* (2010) and Jash and Pan (2007) who indicated that *Trichoderma* was antagonistic to soil borne plant pathogens. *Trichoderma* spp. are known mycoparasites of a number of plant pathogens. *T. harzianum* colonizes *S. rolfsii* hyphae, disrupts mycelial growth and kills the organism. *T. viride* has been shown good in controlling the disease, especially when it is used in combination with fungicide (Surunirajan and Kandhari, 2005).

Agrizeb and Dithane M-45 were found more effective in inhibiting the growth of *S. rolfs*. The present findings agree with the report of Yaqub and Shahzad (2006) that Sancozeb and Dithane M-45 were very effective against the pathogen in the laboratory. In Bangladesh, different fungicides were tested against *S. rolfsii* and it was found that Agrizeb, Sancozeb and Dithane M-45 were effective in reducing disease incidence and severity (Mustika *et al*. 1984). Similar results were observed in soil borne plant pathogens by Babar (1999), Hossain *et al.* (1999) and Yaqub and Shahzad (2006) who reported that carbendazim (as bavistin) and Sancozeb applied to the soil was effective in controlling soil borne diseases. Result of the present study indicated that Haymoxy used against *S. rolfsi* might be excluded from the application schedule and could be replaced by more effective fungicides like Agrizeb and Dithane M-45.

Many workers also reported that the best treatment combines a fungicide that eliminates the soil borne inoculum, a biocontrol agent that possibly takes care of the soil borne inoculum with soil amendment (oil cake) that improve crop productivity by improving nutrients status and soil tilth (Chattopadhyay and Sen, 1996; Anahosur 2001; Basu and Das, 2003).

Using hot water treated seeds of Binatomato-5, superiority of Agrizeb + mustard oil cake + *T. harzianum* was observed as it caused the highest disease reduction of 76.7% over inoculated control. Thus, the best treatment combines a biocontrol agent that possibility takes care of the soil borne inoculum, a fungicide that eliminates the seed inoculum with mustard oil cake conferring added advantage to the system by improving the plant health. This strategy reduces risks of development of resistance in the pathogen to fungicides and requires minimum care by the farmer (Chattopadhyay and Sen, 1996).

The findings of the present integrated approach also have support of the earlier reports of Rahman (2000) and Anahosur (2001) who reported that the soil borne diseases of crops incited by species of *Sclerotium, Rhizoctonia* and *Fusarium* are difficult to be managed through one method of integrated approach *viz*. cultural practices or fungi toxicants or host plant resistance or bio-agents. In recent years efforts were made to reduce environmental effects and rationalize the use of pesticide and manage diseases more effectively which led to the emergence of the new discipline called Integrated Disease Management (IDM) (Anahosur 2001).

## CONCLUSION

Integrated application of management practices i.e. healthy seeds of Binatomato-5 treated with hot water + soil application of Agrizeb 2gm/l @200 ml plant$^{-1}$ + *T. harzianum* (20 g oat plant$^{-1}$) + Mustard oil cake (150 kg/ha$^{-1}$) can reduce disease index of southern stem rot 76.7% and increase yield by 35.7-41.4% under inoculated condition. This system can also be tested for managing stem rot or collar rot of other crops.

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