

EVOLUTION OF DISEASE AND POTENTIAL BIOCONTROL ACTIVITY OF *Trichoderma* SP. AGAINST *Rhizoctonia solani* ON POTATO

POTENCIAL DO BIOCONTROLE POR *Trichoderma* EM RELAÇÃO À EVOLUÇÃO DA DOENÇA CAUSADA POR *Rhizoctonia solani* EM BATATA

Matiar RAHMAN¹; M. Ayub ALI²; Tapan Kumar DEY³; M.M. ISLAM^{4,5}; Laila NAHER⁶; Ahmad ISMAIL⁷

1. Tuber Crops Research Centre, Bogra, Bangladesh; 2. Dept. Plant Pathology, BAU, Mymensingh, 3. Pulse Research Centre, Ishurdi, Pabna, Bangladesh; 4. Department of Forest Management, Faculty of Forestry, University Putra Malaysia, Selangor, Malaysia; 5. Tuber Crops Research Centre, Bangladesh Agricultural Research Institute, Bangladesh; 6. Faculty of Agro-Based Industry, University Malaysia Kelantan, Jeli, 17600, Kelantan. lailanaher@umk.edu.my; 7. Department of Biology, Faculty of Science, University Putra Malaysia, Selangor, Malaysia.

ABSTRACT: Black scurf and stem canker disease cause by the fungal pathogen of *Rhizoctonia solani* and it is an economical important disease of potatoes in Bangladesh and throughout the world. This study evaluated the black scurf and stem canker disease development in potato and antagonistic activity of *Trichoderma* spp. against *R. solani*. The artificial infections were carried out using the inoculums of *R. solani*. The treatments (%inoculum) were: T1 (0% inoculum), T2 (5% inoculum), T3 (10% inoculum), T4 (20% inoculum), T5 (50% inoculum), and T6 (100% inoculum). The infection of stem canker and black scurf on progeny tubers increased with increase in inoculum levels. The highest disease incidence and severity was found in T6 (100% inocula). T6 showed the maximum black scurf infected tubers (russet, deformed and sclerotia). The lowest germination percentage, plant height and tuber yield were also obtained in the same treatment (100% inocula). *Trichoderma* spp reduced the growth of *R. solani* and the highest growth suppression was noted in isolate TM12. According to antagonistic activity, *Trichoderma* spp. reduced the growth of *R. solani* but was not able to stop the pathogen development. This finding showed management of this disease or *R. solani* invasion requires an integrated approach compared to *Trichoderma* single approach.

KEYWORDS: Black scurf, biocontrol, potato. *Rhizoctonia solani*, *Trichoderma* spp, stem canker, hiperparasitismo.

ABBREVIATION: RCBD, randomized complete block design; T, treatment; DAP, days after planting; PDI, percent disease index.

INTRODUCTION

Rhizoctonia solani is a causal fungal pathogen for potato (*Solanum tuberosum* L.), that causes the most serious diseases called stem canker and black scurf, lead to direct reduction to tuber yield and quality. Stem canker consists of stem lesion which involves the reduction of nutrient in the whole plant as a result losses of yield whereas black scurf is the formation of sclerotia, the long term survival of the fungus on newly born tubers (BURPEC; MARTIN, 1992) (Figure 1). The pathogen *Rhizoctonia solani* perpetuates in soil and tubers in the form of sclerotia. As the pathogen is soil inhabitants, so it is difficult to control. Control measures through creation of host resistance have not yet become a viable method because of frequent failure of resistance in the host and development of new races or biotypes of the pathogen.

The use of biological control agent is an attractive approach to combat the disease. Several biocontrol agents were found to use for commercial

formulation and also have demonstrated to control *Rhizoctonia solani* on potato or other diseases. *Trichoderma harzianum* Rifai and *Trichoderma virens* have shown potential to control *Rhizoctonia solani* in pathosystem diseases (BEAGLE-RISTANIO; PAPAVIDAS, 1985; LEWIS; LARKIN, 1997; LEWIS et al. 1998). *Verticillium biguttatum* Gams (JAGER; VELVIS, 1984, 1986), *Pseudomonas fluorescens* Migula (Bagnasco et al., 1998) and *Paenibacillus polymyxa* prazmowski (NIELSEN; SORENSEN, 1997) have shown to control *R. solani* with few are in direct relation to potato diseases and few are in another diseases caused by *R. solani* in different plants. Among many potential antagonistic soil inhabitants, *Trichoderma* spp. has gained considerable importance as a successful bio-control agent for controlling soil borne diseases (CHET; ELAD, 1982).

In Bangladesh, a total of 39 diseases (both biotic and abiotic) of potato have been recorded (ALI; KHAN, 1990). The major soil and tuber-borne diseases are black scurf, stem canker,

bacterial wilt and common scab. Among them, stem canker and black scurf caused by *Rhizoctonia solani* (Kuhn) is the most common and widespread disease throughout the country (ALI; DEY, 1994). *Rhizoctonia* infection can occur on potato plant at any time from sowing to harvest (HIDE et al. 1985; BANVILLE, 1989). Several studies have shown the disease incidence or severity of *R. solani* on potato. Read et al. (1989) reported that the severity of stem canker and black scurf increased with the increase in amount of inoculum. Infection was severe in case of inoculums from sprouted seed. Severe infection due to high amount of inoculum delays shoot emergence decreases the stem height and weight of foliage.

Similarly, Rahman et al. (1996a) observed that the incidence of black scurf of potato increased with the increase in level of inoculums. He also mentioned that the highest level of inocula of *R. solani* caused the maximum reduction in plant growth and tuber yield as well as the highest infection due to wilt of potato. Currently, no report is available in Bangladesh for disease evaluation of stem canker and black scurf of potato. Therefore, the present study was developed to evaluate the stem canker and black scurf diseases of potato as well as biocontrol activity of *Trichoderma spp.* against *R. solani* (Figure 2).

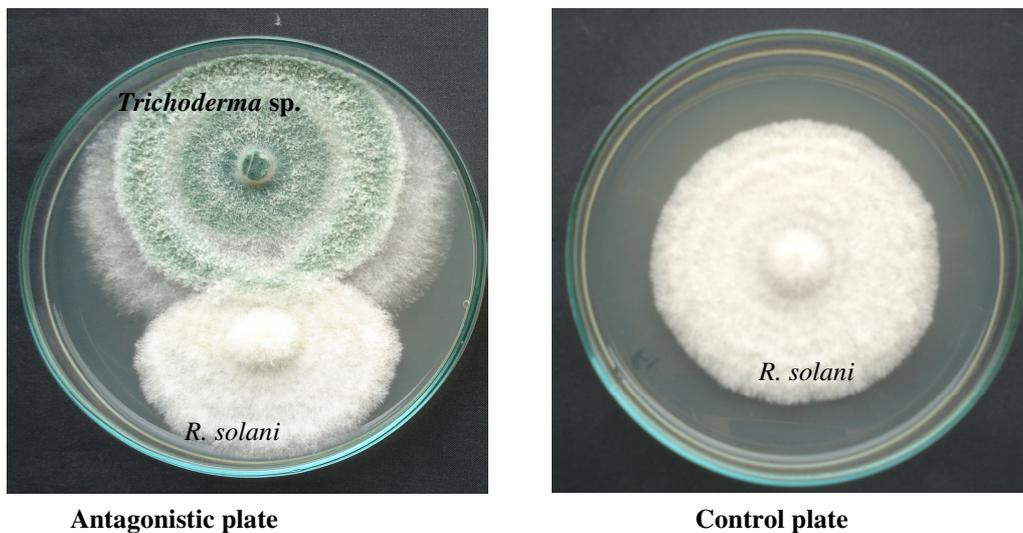


Figure 2. Comparative colony diameter of *Trichoderma sp.* and *Rhizoctonia solani*.

MATERIAL AND METHODS

Field study and Experimental site and soil characteristics

The experiment was conducted at Tuber Crops Research Sub-Centre (TCRSC), BARI, during 2007-2008. The soil is sandy loam in texture with p^H 5.6. The total rainfall during the cropping season (November-April) was 65, 25 and 25 mm in 2007-08, 2008-09 and 2009-10, respectively. The soil was prepared as a good tith. The weeds and stubbles were removed from the field. Finally the soil was leveled with a harrower (laddering) by a power tiller.

Treatment details, fertilizer and manure application

The experiment was consisted with six treatments such as T_1 (0% inocula used as control), T_2 (5% inocula), T_3 (10% inocula), T_4 (20% inocula), T_5 (50% inocula) and T_6 (100% inocula). The experiment was laid out in a randomized

complete block design (RCBD) with four replications. Cowdung and chemical fertilizers were applied to supply nutrients to the crop. Urea, triple superphosphate (TSP), muriate of potash (MoP) and gypsum were used as sources of N, P, K and S, respectively. Entire quantity of cowdung, Phosphorous, Sulphur, Zinc and half of nitrogen and potassium were applied before planting and mixed into the soil. Remaining nitrogen was side-dressed at 30 DAP (days after planting) at the time of earthing up.

Planting and harvesting

Potato variety Diamant (*Solanum tuberosum*) was used as a test crop. The infected and uninfected seed tubers of Potato were collected from Breeder Seed Production Centre (BSPC) Debigong, Bangladesh. The unit plot size was 3 m × 3 m. Tubers of potato were planted with a spacing of 60 cm × 25 cm on 22 November in each year. Potato was harvested on 20 February.

Intercultural operation

Intercultural operations such as weeding and mulching were done as and when required. Irrigations were given four times during the whole growing season. First time, light irrigation was applied at 7 days after planting (DAP) to ensure proper germination. Second irrigation was given at 30 DAP followed by earthing up and side-dressing (urea fertilizer). Third and fourth irrigations were applied at 48 and 63 DAP, respectively. Dursban (0.5%) and Admire (0.1%) were applied to control cutworm and aphid, respectively. Secure (0.1%) was sprayed at 10 days interval to prevent late blight disease of potato.

Data collection

The germination per cent, number of stems per hill and plant height were recorded at 30, 50 and

60 DAP, respectively. The yield data were noted at harvest. The disease incidence (%) was assessed at 70 DAP. Twenty plants were randomly selected in each plot and uprooted carefully from soil, washed with water and investigated for infection. Numbers of infected and healthy plants were counted in each plot and incidence percentage was calculated using equation (1).

$$\text{Incidence (\%)} = \frac{\text{Infected plants}}{\text{Healthy plants}} \times 100 \dots (1)$$

Per cent Disease Index (PDI)

Per cent severity index was also noted at 70 DAP. Twenty plants were collected and washed in same procedure then plants were investigated individually and categorized into different group according to the grade of infection using the following 0-6 scale (DEY, 2010) (Box 1).

Box 1. Grade of infection of *Trichoderma spp*

Grade	Description
0	No symptom on stolon
1	Minute brown lesion on stolon or root
2	Moderately brown lesion on stolon and curling tendency on central leaf
3	Stolon symptom discolored accompanied by brown discoloration on roots
4	Brown to black discoloration on underground parts, tissue discoloration and tissue squeezed/ curling of growing leaves.
5	Profuse emerging of auxiliary leaves, leaf size, reduced markedly and pale green on leaf margin.
6	Production of aerial tuber with green colour

The number of stem canker infected plants under each grade (0-6 scale) was recorded and the percent disease index (PDI) was calculated by the equation (2); $\text{PDI} = \frac{\text{Class frequency}}{\text{No. of plants assessed}} \times \text{highest score} \times 100 \dots (2)$

Tuber infection

The black scurf infected tubers were separated into three groups such as russet, deformed and sclerotia (CHAND; LOGA 1982). The number of tubers under each group was counted and the respective weight was recorded. Number and weight of healthy tubers of each plot was also noted.

Statistical analysis

The analysis of variance for different parameters (germination, plant height, stems hill⁻¹, yield, disease incidence, severity and infected seed) were done following the ANOVA test and the mean values were compared by DMRT (P=0.05) (STEEL; TORRIE, 1960).

Determination of biocontrol potential of *Trichoderma spp.* against *Rhizoctonia solani*

Trichoderma spp. isolates TM3, TM6, TM11, TM12, TM14 were collected from Plant Pathology Division Laboratory, Bangladesh

Agricultural Research Institute, Gazipur, Bangladesh.

The stems of buck wheat (*Fagopyrum esculentum* Moench.) were used as bait for isolation of *R. solani* from soil. First, the stems were dried which previously collected from Plant Genetic Resource Center, BARI, Gazipur. The materials were preserved for future use. Next, the stems were cut into 3-4 cm small pieces and autoclave at 121⁰ C for two hours. 5-6 pieces were buried at depth of 6 cm in a pit field soil. After 3-5 days of incubation the pieces of bait were recovered from the soil, washed in running tap water and then rinsed three times with distilled water, dipped into 200-ppm streptomycin sulphate solution for 10 minutes and blotted with sterilized blotting paper. The baits were further cut into small pieces. The baits were plated (2 bits/ plate) on 1.5% water agar (WA) medium in glass petridishes. About 0.2 g streptomycin sulfate and 0.1g metalaxy1 was added with 1 L of WA medium. The plates were incubated in darkness at 25 ± 1⁰ C for 48 hours. The hyphal tips of mycelial

growth of *Rhizoctonia solani* were transferred on the PDA glass petridish plate and incubated for 48 hours. A bit of pure culture was placed at the middle of new Petridishes. The plates were incubated for five days. The plates having full growth of *R. solani* were used for the following experiment.

Efficacy of *Trichoderma* spp.

The *in-vitro* antagonism of *Trichoderma* isolates and *Rhizoctonia solani* were tested on PDA by dual culture plate technique. Mycelial blocks (5 mm) were cut from the periphery of 3 days old culture of both *Trichoderma* isolates and *Rhizoctonia solani*. Two mycelial blocks, one from *Trichoderma* isolates and other from *Rhizoctonia solani* were placed on PDA plate, facing opposite to each other and incubated at $25^{\circ} \pm c$. The treatments were arranged following CRD with four replications. The radial growth of each colony was measured at 24 and 72 hour interval.

RESULTS AND DISCUSSION

Effect of inocula of *Rhizoctonia solani* on germination and growth parameters

The levels of inoculum of *Rhizoctonia solani* had significant effect on the germination (%)

of the tubers. The highest germination (98.33%) was found in the control, which was significantly higher than the other treatments. The second highest germination (94%) was recorded in T2 followed by T3 and the lowest was in T6 (Table 1). It was observed that the germination percentage increased with increase in tuber borne inocula levels. HIDE et al. (1985) reported that potato tubers are mostly damaged by *Rhizoctonia solani* infection, leading to inhibition of germination and killing of underground sprouts thus delaying emergence. Similar results were also found by Banville (1989). The number of stem hill⁻¹ and plant height was also varied due to different treatments but their effects were not significant (Table 1). The stem hill⁻¹ ranged from 4.43 to 5.10, having the highest in T1 followed by T4 (4.83). The minimum stem hill⁻¹ (4.43) was noted in T2 (Table 1). In treatment T1 showed the highest plant height (60.6 cm) followed by T2 (59.6 cm) and T3 (58.3 cm). Read et al. (1989) also observed that severe infection due to high amount of inoculum delayed shoot emergence and decreased the plant height but did not affect the number of stems plant⁻¹ which are in agreement with the findings of our study.

Table 1. Effect of tuber-borne inoculum of *R. solani* on germination and growth parameters of potato

Tuber-borne Inocula	Germination (%)	No. of stem / hill (NS)	Plant height (cm) (NS)
T ₁ =0 % inocula	98.33 a	5.1	60.58
T ₂ =5% inocula	94.08 b	4.43	59.55
T ₃ =10% inocula	90.49 c	4.65	58.25
T ₄ =20% inocula	86.33 d	4.83	57.88
T ₅ =50% inocula	84.99 bd	4.70	56.08
T ₆ =100% inocula	80.95 e	4.48	54.33
CV%	0.94	9.64	7.27

Means followed by the same letter within a column did not differ significantly at the 5% level by DMRT. NS = Not significant

Effect of inocula of *Rhizoctonia solani* on the incidence and severity of stem canker

The effect of tuber-borne inoculum levels of *Rhizoctonia solani* on the incidence and severity of stem canker of potato in the field are shown in Table 2 and Figure 1. The incidence and severity were significantly variable among the treatments. The

maximum incidence (51.24%) was found in T₆, which was significantly higher than the other treatments. T₅ showed the second highest incidence (43.3%) and the minimum (18.33%) was in the control (Table 2). A percent disease index (PDI) of stem canker was also significantly influenced by the treatments. The highest PDI (21.25%) was found in

T6 which was also significantly higher than the other treatments. Treatments T3 and T4 showed statistically similar PDI. The lowest PDI (7.29%) was recorded in the control. Read et al. (1989) applied different amount of inoculum of *R. solani* to sprouted seed tubers during planting, the severity of stem canker increased with increase in inoculum levels. The infection was also severe in case of

inoculum from sprouted seed. Naz et al. (2008) conducted an experiment with three levels of inoculum such as 10, 15 and 20 g of *R. solani* on the development of stem canker and observed that the disease incidence increased with increase in inoculum level of *R. solani* which corroborated our results.

Table 2. Effect of tuber-borne inoculum of *R. solani* on the incidence of stem canker of potato

Tuber-borne Inocula	Incidence (%)	Severity (PDI)
	18.33 e	7.29 e
T ₁ =0 % inocula	27.08 d	9.16 d
T ₂ =5% inocula	33.33 c	12.08 c
T ₃ =10% inocula	38.33 bc	13.75 c
T ₄ =20% inocula	43.33 b	16.87 b
T ₅ =50% inocula	51.24 a	21.25 a
T ₆ =100% inocula	4.91	6.67
CV%		



Figure 1. Different types of disease symptoms on potato tuber in pathogenicity by *R. solani*.

Effect of inocula of *Rhizoctonia solani* on the black scurf infected tubers

Tuber borne inoculum played significant role in production of different types of symptoms of black scurf of potato (Table 3 and Figure 1). *R. solani* produced three types of symptoms such as russet, deformed and *sclerotia* on the tubers. The highest number of russet (20 plot⁻¹) and deformed (17.0 plot⁻¹) tubers were recorded in T6, which was significantly higher than the other treatments (Table 3). T5 showed the second highest number of russet (17.3 plot⁻¹) and deformed (15.0 plot⁻¹) tubers. The minimum number of russet (8.25 plot⁻¹) and deformed (5.50 plot⁻¹) tubers were noted in the control (Table 3). There was a large variation in the number of *sclerotia* bearing tubers among the treatments. The maximum number of *sclerotia*

bearing tubers (90.5 plot⁻¹) was obtained in T6 which was significantly different from all other treatments. T5 showed the second highest number of *sclerotia* bearing tubers (60.5 plot⁻¹) and the lowest (4.00 plot⁻¹) was noted in the control. Soil-borne inoculum of *R. solani* is the main cause of black scurf on potato tubers and also contributes to eyes germination inhibition, sprouts killing, stem, stolon and root damage (HIDE et al.1973; FRANK; LEACH, 1980). Black scurf attacks potato plants causing delay in tuber initiation and reduction in tuber yield. *Sclerotia* present in harvested tubers reduce market value of the crop (THRIMULACHAR, 1953; COTHER, 1979). RAHMAN et al. (1996a) reported that the incidence of black scurf of potato increased with increase in level of *Rhizoctonia solani* inoculum.

Table 3. Effect of tuber-borne inoculum of *Rhizoctonia solani* on the number of black scurf infected tubers

Tuber borne Inocula	Number of infected tuber / plot		
	Russet	Deformed	Sclerotia
T ₁ =0 % inocula	8.25 d	5.50 d	4.00 f
T ₂ =5% inocula	12.75 c	9.75 bcd	18.00 e
T ₃ =10% inocula	13.75 c	10.75 cd	24.75 d
T ₄ =20% inocula	14.45 c	11.00 c	44.50 c
T ₅ =50% inocula	17.25 b	15.00 b	60.50 b
T ₆ =100% inocula	20.00 a	17.00 a	90.50 a
CV%	14.89	16.74	9.28

Means followed by the same letter within a column did not differ significantly at the 5% level by DMRT.

The number of infected tubers plot⁻¹ was significantly variable among the treatments (Table 3). T6 showed the maximum infected tubers (20, 17 and 90.5 plot⁻¹ for russet, deformed and *sclerotia*, respectively), which was significantly higher than the other treatments. The minimum infected tubers (8.25, 5.50 and 4.00 plot⁻¹ for russet, deformed and *sclerotia*, respectively) were noted in the control (Table 3). The tuber-borne inoculum showed significant variation in the weight of tubers having different type of symptoms (Table 4). The highest weight of russet tubers (1370 g plot⁻¹) was recorded in T6 which was identical to T5 (1250 g plot⁻¹) but significantly higher than the other treatments. The

minimum weight of russet tubers (600 g plot⁻¹) was noted in the control (Table 4). The maximum weight of deformed (1200 g plot⁻¹) and *sclerotia* bearing (6700 g plot⁻¹) tubers were recorded in T6 which was also significantly higher than the other treatments. T5 showed the highest weight of deformed and *sclerotia* bearing tubers and the lowest weight of deformed (515 g plot⁻¹) and *sclerotia* bearing (200 g plot⁻¹) tubers were noted in the control (Table 4). It was observed that the weight of infected tubers increased with increase in inocula percentage. But reverse is true in case of healthy tubers.

Table 4. Effect of tuber-borne inoculum of *R. solani* on the weight of black scurf infected tubers

Tuber-borne Inocula	Weight of infected tuber/plot (g)		
	Russet	Deformed	Sclerotia
T ₁ =0 % inocula	600 d	515 e	200 f
T ₂ =5% inocula	900 c	617 d	1090 e
T ₃ =10% inocula	970 c	820 c	1170 d
T ₄ =20% inocula	998 c	850 c	3110 c
T ₅ =50% inocula	1250 a	920 b	4320 b
T ₆ =100% inocula	1370 a	1200 a	6700 a
CV%	14.54	23.36	9.94

Means followed by the same letter in the same column did not differ significantly at the 5% level by DMRT.

Effect of inocula of *Rhizoctonia solani* on the yield of potato

The potato yield was also significantly influenced due to different treatments (Table 5, Figure 3). It ranged from 17.6 to 21.5 t ha⁻¹, the highest yield (21.5 t ha⁻¹) was found in the control followed by T2 (19.9 t ha⁻¹) and T3 (19.1 t ha⁻¹). The minimum potato yield (17.6 t ha⁻¹) was obtained in

T6. Rahman *et al.* (1996 b) reported that the highest level of inocula caused the maximum reduction in plant growth and tuber yield and the highest infection. Yield loss increased with increase in disease intensity; 50% incidence of seed tubers reduced germination by 11% and yield by 68.2% (Lakra, 1992).



Figure 3. Yield production of potato tuber as influenced by different level of *R. solani* inocula.

Table 5. Effect of inoculum levels of *R. solani* on tuber yield of potato

Tuber-borne Inocula	Yield (tha ⁻¹)
	21.50 a
T ₁ =0 % inocula	19.94 ab
T ₂ =5% inocula	19.11 ab
T ₃ =10% inocula	18.86 ab
T ₄ =20% inocula	18.50 b
T ₅ =50% inocula	17.59 b
T ₆ =100% inocula	8.91
CV%	

Means followed by the same letter within a column did not differ significantly at the 5% level by DMRT.

Determination of biocontrol potential of some isolates of *Trichoderma* spp. against *Rhizoctonia solani*

The antagonistic effect of some isolates of *Trichoderma* spp. on the vegetative growth of pathogenic fungus *Rhizoctonia solani* is shown in Table 6. The study revealed that all the isolates of *Trichoderma* remarkably reduced the radial growth of *R. solani* on PDA. The isolate TM12 suppressed 15.63% vegetative growth of *R. solani* at 48 hours after inoculation while TM11 and TM6 isolates reduced growth 2.08 and 6.25%, respectively. After 72 hours all *Trichoderma* isolates showed better performance to reduce the growth of *R. solani* except TM14. Isolate TM12 showed the highest growth suppression (32.2%) followed by TM6 (29.4%) and TM11 (28.9%). The minimum suppression (7.50%) was noted in TM14. The *Trichoderma* isolates are antagonistic of *R. solani* on PDA media. They inhibited the vegetative growth of *Rhizoctonia solani* (26.66-32.23%) but none of them was able to stop the growth of the

pathogen. *Trichoderma* spp. is the most biocontrol agent to manage the plant diseases. Thus, this study measured the antagonistic activity of *Trichoderma* against *R. solani* in plate assay (Figure 2). The result showed that the isolated of *Trichoderma* spp. was able to slow the growth of *R. solani* but could not manage to stop the pathogen growth. This study was a trial to evaluate the antagonistic activity of *Trichoderma* spp but the result was not similar with other study, for example *Trichoderma* slightly control of black scurf of potato caused by *R. solani* and when used *Bacillus subtilis* the control activity was increased compared to *Trichoderma* alone (BREWER; LARKIN 2005). The reason is not clear that why *Trichoderma* could not manage to stop the growth of *R. solani*, or it can be that the media or PDA was not enough nutrient supporters for the growth of *Trichoderma* spp. to suppress the *R. solani* (Figure 2). A previous study found that the antagonistic activity of *Trichoderma* increased in combined media compared to PDA alone (RODRIGUES et al. 2009).

Table 6. Suppression of vegetative growth of *Rhizoctonia solani* by different isolates of *Trichoderma* after 48 and 72 hours of inoculation

Isolate number	Radial growth of <i>Rhizoctonia solani</i> after 48 hrs. (cm)		Growth suppression (%)	Radial growth of <i>Rhizoctonia solani</i> after 72 hrs. (cm)		Growth suppression (%)
	Dual culture	Mono culture		Dual culture	Mono culture	
TM 3	3.53	3.84	8.07	4.21	5.74	26.66
TM 6	3.60	3.84	6.25	4.05	5.74	29.44
TM 11	3.76	3.84	2.08	4.08	5.74	28.92
TM 12	3.24	3.84	15.63	3.89	5.74	32.23
TM 14	3.46	3.84	9.90	5.31	5.74	7.49

TM= *Trichoderma* isolate

CONCLUSIONS

There were significant variation among the inoculum levels of *Rhizoctonia solani* on the stem canker and black scurf in progeny tubers.

The infection of stem canker and black scurf on progeny tubers increased with increase in inoculum levels.

The highest disease incidence and severity was found in T6 treatment. T6 showed the maximum black scurf infected tubers (russet, deformed and sclerotia).

The lowest germination percentage, plant height and tuber yield were also obtained in the same treatment. *Trichoderma* spp reduced the growth of *R. solani* and the highest growth suppression was noted in isolate TM12.

The future study is needed to compare the antagonistic activity in different types of nutrient amendment and the integrated approach should be carried out which may be helpful to control stem canker and black scurf disease in potato.

RESUMO: A rizoctoniose ou crosta negra causada por *Rhizoctonia solani* é a mais importante doença nos campos de batata em Bangladesh, bem como em várias regiões do mundo. Este trabalho avaliou o potencial do biocontrole com *Trichoderma* spp. e sua ação antagonista contra *R. solani* em batateira. Realizou-se as avaliações do potencial antagonista usando inoculação artificial de *R. solani*. Os tratamentos (% de inóculo) foram: T1 (0% de inóculo), T2 (5%),

T3 (10%), T4 (20%), T5 (50%) , e T6 (100% de inóculo). A infecção de rizoctoniose na haste e crosta negra nos tubérculos aumentou proporcionalmente com o aumento do nível de inóculo. A maior incidência e severidade da doença ocorreu no tratamento 6 (100 % de inóculo), o qual apresentou maior quantidade de tubérculos infectados e deformados com escleródios em sua superfície. A menor porcentagem de germinação e produção de tubérculos também foi encontrada no tratamento 6, o qual também apresentou menor altura de planta. *Trichoderma* spp reduziu o crescimento de *R. solani* e a maior atividade de supressão do crescimento foi encontrada pelo isolado TM12. Foi detectada a atividade antagonista de *Trichoderma* spp. em reduzir o crescimento de *R. solani*, mas este não inibiu o crescimento total do patógeno. Conclui-se que o manejo da rizoctoniose da batateira por colonização de *R. solani* necessita táticas de manejo integrado em detrimento do uso isolado do manejo ou biocontrole com *Trichoderma* spp.

PALAVRAS-CHAVE: Manejo da rizoctoniose. Cancro da haste por rizoctoniose. Biocontrole. Hiperparasitismo. Batateira. *Trichoderma*. *Rhizoctonia solani*.

REFERENCES

- ALI, M. S., DEY, T. K. Pathological research on tuber crops in Bangladesh. *In: Proc. Workshop on Tubers Crops on Transfer of Technology of CDP crops under Research Extension Linkage Programme.*, p. 159-165. 1994.
- BANVILLE, G. J. Yield losses and damage to potato plants caused by *Rhizoctonia solani* Kuhn. *Am. Potato J.*, Canada, v. 66, p. 821-834. 1989.
- BEAGLE-RISTANIO J. E., PAPAVIDAS, G. C. Biological control of *Rhizoctonia* stem canker and black scurf of potato. *Phytopathol. America*, v. 75, p. 560-564. 1985.
- BREWER, M. T., LARKIN, P. R. Efficacy of several potential biocontrol organisms against *Rhizoctonia solani* on potato. *Crop Protect.*, v. 24, p. 939-950. 2005.
- CHAND, T., LOGAN, C. Reaction of ten potato cultivars to stem canker and black scurf of potato caused by *Rhizoctonia solani*. *Ann. Appl. Biol.*, v. 100, p. 102-103. 1982.
- CHET, I.; ELAD, Y. Prevention of infection by bio-logical means. *In. La Selection des plantes*, Bor-deaux (France). *Colloqua I'INRA*. France, v. 11, p. 95-205. 1982.
- COTHER, E. J. Presence of *Rhizoctonia solani* kuhn in nature. Cultivars and its implications for future potato growing. *Australas. Plant Pathol.*, Australia, v. 8, p. 14-15. 1997.
- DEY, T. K. Tuber crops diseases-their identification, method of recording, rating scale and grading system. *Oil seed Res. Center of Bangladesh Agr.*, Bangladesh, p. 28-31. 2010.
- FRANK, J. A., LEACH, S. S. Comparison of tuber-borne and soil-borne inoculum in the *Rhizoctonia* disease of potato. *Phytopathol.*, v. 70, p. 51-53. 1980
- HIDE, G. A., READ, P. J., SANDISON, J. P. Stem canker (*Rhizoctonia solani*) of maincrop potatoes. *Ann. Appl. Biol.*, v. 106, p. 423-437. 1985.
- HIDE, G. A., HIRST, J. M., STEDMAN, O. J. Effects of black scurf (*Rhizoctonia solani*) on Potatoes. *Ann. Appl. Biol.*, v. 74, p. 139-148. 1973.
- JAGER, G.; VELVIS, H. Biological control of *Rhizoctonia solani* on potatoes by antagonists. 2. Sprout protection against soil-borne *R. solani* through seed inoculation with *verticillium biguttatum*. *Neth. J. Plant Pathol.*, Netherlands, v. 90, p. 29-33. 1984.

JAGER, G.; VELVIS, H. Biological control of *Rhizoctonia solani* on potatoes by antagonists. 5. The effectiveness of three isolates of *Verticillium biguttatum* as inoculums for seed tubers and of a soil treatment with a low dosage of pencycuron. **Neth.J. Plant Pathol.**, Netherlands, v. 92, p. 231-238. 1986.

LENHTONEN, M.; WILSON, P. S.; AHVEBBIEMI, P.; VALKONENE, P. T. J. (2009)

Formation of canker lesions on stems and black scurf on tubers in experimentally inoculated potato by isolates of AG2-1, AG3 and AG5 of *Rhizoctonia solani*: a pilot study and literature review. **Agr. food Sci.**, v. 18, p. 223-233.

LEWIS, J. A.; LARKIN, R. P. Extruded granular formulation with biomass of biocontrol *Gliocladium viren* and *Trichoderma* spp. to reduce damping off of egg plant caused by *Rhizoctonia solani* and saprophytic growth of the pathogen in soil-less mix. **Biocontrol Sci.Tech.**, v. 7, p. 49-60. 1997.

LEWIS, J. A.; LARKIN, R. P. Formulation of the biocontrol fungus *Cladorrhinum foecundissimum* to reduce damping off diseases caused by *Rhizoctonia solani* and *Pythium ultimum*. **Biological control**, v. 12, p. 182-190. 1998.

MUTHUKUMAR, A., BHASKAVAN, R. Efficacy of anti-microbial metabolites of *Pseudomonas fluorescens* (Trevisan) Migula against *Rhizoctonia solani* Kuhn. and *Pythium* sp. **J. Biol. control**, v. 21, p. 105-110. 2007.

NAZ, F.; RAUF, C. A.; ABBASI, N. A.; HAQUE, I. U.; AHMAD, I. Influence of inoculum levels of *Rhizoctonia solani* and susceptibility on new potato germplasms. **Pak. J. Bot.**, Pakistan, v. 40, p. 2199-2209. 2008.

NIELSEN, P.; SORENSEN, J. (1997) Multi-target and medium independent fungal antagonism by hydrolytic enzymes in *Paenibacillus polymyxa* and *Bacillus pumilus* strains from the barley rhizosphere. **FEMS Microbiol. Ecol.**, v. 22, p. 183-192.

READ, P. J.; HIDE, G. A., FIRMAER J. P.; HALL, S. M. (1989.) Growth and yield of potatoes as affected by severity of stem canker (*Rhizoctonia solani*). **Potato Res.**, European, v. 32, p. 9-15.

RAHMAN, M. L.; HOSSAIN, M. M.; ASHRAFUZZAMAN, M.; ISLAM, T. Effect of inoculum levels of *Rhizoctonia solani* on the incidence of black scurf disease of potato. **Bangladesh J. Plant Pathol.**, Bangladesh, v. 12, p. 21-22. 1996

RODRIGUES, R. C. B., BRAGA, B. R. M., TORNISIELO, T. M. S., CARMONA, C. E., ARRUDA, M. V., NETTO, C. J. (2009) Comparative growth of *Trichoderma* strains in different nutritional sources, using bioscreen C automated system. **Braz. J. Microbiol.**, São Paulo, v. 40, p. 404-410.

Thirumalachar, M. J. (1953) *Rhizoctonia solani* infections of potato tubers in India. **Phytopathol.**, v. 43, p. 645-647.