EVALUATION OF *TRICHODERMA* BASED BIOPESTICIDE FORMULATION FOR CONTROLLING DAMPING OFF PATHOGEN OF EGGPLANT AND TOMATO SEEDLING

MD. JANNATUL ADAN

DEPARTMENT OF PLANT PATHOLOGY

SHER-E-BANGLA AGRICULTURAL UNIVERSITY

DHAKA-1207, BANGLADESH

DECEMBER, 2013

EVALUATION OF *TRICHODERMA* BASED BIOPESTICIDE FORMULATION FOR CONTROLLING DAMPING OFF PATHOGEN OF EGGPLANT AND TOMATO SEEDLING

BY

MD. JANNATUL ADAN

REGISTRATION NO. 08-02677

A thesis

Submitted to the Faculty of Agriculture
Sher-e-Bangla Agricultural University, Dhaka,
In partial fulfilment of the requirements
For the degree of

MASTER OF SCIENCE (MS)

IN

PLANT PATHOLOGY

SEMESTER: July – December, 2013

Approved by:

(Prof. Dr. Md. Rafiqul Islam) **Department of Plant Pathology** Sher-e-Bangla Agricultural University Supervisor	**(Prof. Nazneen Sultana)** **Department of Plant Pathology** Sher-e-Bangla Agricultural University Co-supervisor

(Prof. Dr. F. M. Aminuzzaman)
Chairman
Department of Plant Pathology
Sher-e-Bangla Agricultural University
Dhaka- 1207

DEDICATED

TO

MY BELOVED

PARENTS

ACKNOWLEDGEMENTS

All praises to “Almighty Allah” whose blessings enabled the author for successful completion of this research work.

The author sincerely expresses his heartfelt gratitude, great pleasure, immense indebtedness and sincere appreciation with deepest emotion to his honourable teacher and research supervisor Prof. Dr. Md. Rafiqul Islam, Department of Plant Pathology, Sher-e-Bangla Agricultural University, Dhaka for his scholastic guidance, precious suggestions, affectionate feelings, constant encouragement and constructive criticisms and inspirations during the whole period of the research work and preparing of the manuscript.

The author extends his deep sense of gratitude and indebtedness to his honourable teacher and co-supervisor Prof. Nazneen Sultana, Department of Plant Pathology, Sher-e-Bangla Agricultural University, Dhaka for her kind inspiration, suggestions and guidance for entire period of the research work.

The author desires to express his respect and deepest sense of gratitude to all the respectable teachers of the Department of Plant Pathology, Sher-e-Bangla Agricultural University, Dhaka for their valuable suggestions and kind co-operation during the period of the study.

The author desires to express his heartiest gratitude to Afsana Jahan, MS student, Department of Plant Pathology, Sher-e-Bangla Agricultural University, Dhaka for her inspirations and best co-operation during the research period and preparation of the thesis.

Cordial appreciation and thanks are extended to all the staff of Plant Pathology Laboratory and farm workers of Sher-e-Bangla Agricultural

University, for their assistance and co-operation for conducting the research work.

Finally, the author wishes to acknowledge his heartfelt gratitude and profound respect to his beloved parents and all other relatives for their blessing and inspiration throughout the entire period of his study.

The author

CONTENTS

RESULTS (Contd.)

LIST OF TABLES

LIST OF PLATES

LIST OF PLATES (Contd.)

LIST OF FIGURES

LIST OF APPENDICES

EVALUATION OF *TRICHODERMA* BASED BIOPESTICIDE FORMULATION FOR CONTROLLING DAMPING OFF PATHOGEN OF EGGPLANT AND TOMATO SEEDLING

ABSTRACT

The effect of nine *Trichoderma* based substrates viz. T_1 (*Trichoderma* + Rice bran + Peat soil + Water), T_2 (*Trichoderma* + Wheat bran + Peat soil + Water), T_3 (*Trichoderma* + Lentil bran + Peat soil + Water), T_4 (*Trichoderma* + Gram bran + Peat soil + Water), T_5 (*Trichoderma* + Black gram bran + Peat soil + Water), T_6 (*Trichoderma* + Mustard oil cake + Peat soil + Water), T_7 (*Trichoderma* + Grass Pea bran + Peat soil + Water), T_8 (*Trichoderma* + Saw Dust + Peat soil + Water), T_9 (Control) were evaluated for sporulations of *Trichoderma harzianum* and acting against *Sclerotium rolfsii* for the management of damping off of eggplant and tomato seedlings. The effect of the treatments varied significantly in terms of production of *Trichoderma* spore and reducing damping off and tip over, increasing germination percentage, plant height, seedling vigor and fresh weight of vegetable seedlings in comparison to control. Among the treatments soil application with T_5 (*Trichoderma* + Black gram bran + Peat soil + Water), T_7 (*Trichoderma* + Grass Pea bran + Peat soil + Water) and T_4 (*Trichoderma* + Gram bran + Peat soil + Water) showed the promising effect in controlling pre-emergence damping off, post-emergence damping off, and tip over and of increasing germination percentage, plant height, vigor index and fresh weight of seedlings. The highest germination was observed in treatment T_5 in eggplant 78.00% (16 DAS) and in tomato 83.15% (13 DAS). The lowest (6.33% and 3.33%) post-emergence damping off was observed in T_5 in eggplant and tomato seedling at 16 DAS and 13 DAS respectively. The lowest pre- emergence damping off and tip over were observed in T_5 in eggplant (2.33% and 1.33%) and in tomato seedlings (1.85% and 0.55 %).

DEPARTMENT OF PLANT PATHOLOGY

Sher-e-Bangla Agricultural University

Sher-e-Bangla Nagar, Dhaka-1207

Phone: 9134865

CERTIFICATE

*This is to certify that the thesis, entitled "**EVALUATION OF TRICHODERMA BASED BIOPESTICIDE FORMULATION FOR CONTROLLING DAMPING OFF PATHOGEN OF EGGPLANT AND TOMATO SEEDLING**" submitted to the Faculty of Agriculture, Sher-e-Bangla Agricultural University, Dhaka in the partial fulfilment of the requirements for the **MASTER OF SCIENCE IN PLANT PATHOLOGY,** embodies the result of a piece of bonafide research work carried out by **MD. JANNATUL ADAN,** Registration No. 08-02677, under my supervision and guidance. No part of this thesis has been submitted for any other degree in any other institutes.*

I further certify that any help or sources of information received during the course of this investigation have been duly acknowledged.

Dated:

Dhaka, Bangladesh

_____

(Prof. Dr. Md. Rafiqul Islam)

Department of Plant Pathology

Sher-e-Bangla Agricultural University

INTRODUCTION

Vegetables play an important role in balance diet of human beings. Vegetables are rich sources of vitamins and minerals and also a good source of carbohydrates. Vegetables of Bangladesh are grouped into summer, winter and year round on the basis of growing season. Total production of vegetables meets up to 45-50% of the requirement of the country. Eggplant, tomato and chilli are important high value crops among vegetables in Bangladesh. These are common and economically important vegetables in Bangladesh (BBS, 2008).

The eggplant or brinjal (*Solanum melongena* L.) belong the family Solanaceae (also known as the Aubergine) and genus *Solanum*. It is grown in Bangladesh, India, China, Pakistan and the Philippines. It is also a popular vegetable crop in France, Italy, USA and the Mediterranean and Balkans areas (Bose and Som, 1986). Brinjal used as a popular cooked vegetable item worldwide including most of the Asian countries and in many European and American countries. It is widely grown all over the world including tropical, subtropical and temperate region. Being a subtropical country it is widely grown in Bangladesh both in Rabi and kharif seasons (Haque, 2006). Brinjal is the second most important vegetable crop next to potato in Bangladesh in respect of acreage and production (BBS, 2011). The total area of brinjal cultivation was 78000 acres where 44000 acres was grown in Kharif season (summer) and 34000 acres in Rabi season (winter) with a total annual production of 246000 tons (BBS, 2011), which is very low in comparison to that of other countries like India, China, Egypt etc.

Such a potential crop is known to suffer from twelve diseases. Among them damping off and foot rot caused by *Sclerotium rolfsii* has been treated as one of the major constrains of eggplant cultivation (Klean and Murugesan, 2003).

Tomato *(Lycopersicon esculentum Mill)*, a member of the family Solanaceae, is the most popular vegetable in the world because of its taste, colour and high nutritive value and also for its diversified uses (Bose and Sam, 1986). In Bangladesh, tomato is cultivated mainly in homestead gardens as well as in fields during winter and in limited scale at summer season. Tomato was grown in 61000 acres of land in Bangladesh, the total production was 232000 tons in 2010-2011 (BBS, 2011). About 85% of total tomatoes are grown in six grater districts, namely Comilla, Dhaka, Jessore, Chittagong, Sylhet and Rajshahi. As a 100% edible vegetable tomato plays a vital role in human nutrition having 15% vitamin and it is 100% edible (Gowda and Kaul, 1982).

Per acre yield of tomato was 3798 kg in 2010-2011 (BBS, 2011) which was very low compared to other countries. Diseases are main constrains to lower yield of tomato in Bangladesh which cause about 30-40% yield loss of this crop annually (Anonymous, 1992). Among the various diseases, *Sclerotium rolfsii, Fusarium oxysporum,* and *Rhizoctonia solani* causing damping off of seedling is the most prevalent in the tomato growing areas in Bangladesh. The disease has also been reported in many countries in the world (Nene *et al*., 1996).

Damping off is a serious disease of vegetables grown in nursery bed. The most common fungi reported to be responsible for damping off are *Pythium* sp., *Fusarium oxysporum, Sclerotium rolfsii, Phytophthora* sp. and *Rhizoctonia solani* etc. (Sing, 1984; Ahmed and Hossain, 1985).

The damping off occures in nursery bed as pre-emergnbce damping off and post-emergence damping off. In pre-imergence damping off, the young seedlings are killed beofere they emerged from the soil while im post-emergence damping off, the infected seedlings are topple down due to infection in the coller reagion after the emergence of seedlings from the soil.

The fungi *Fusarium oxysporum*, *Sclerotium rolfsii* and *Rhizoctonia solani* are soil inhabiting pathogen with wide host range and therefore, very difficult to control them.(Rangswami, 1988; Talukder, 1974; Sing, 1984; Elango, 1986; Das, 1984; Martin and Torres, 1989).

There are several methods for controlling damping off disease of seedling. Farmers try to overcome this problem through different cultural practices and use of chemical fungicides. But the control of soil borne pathogens with chemicals is very expensive and is almost impractical in Bangladesh. In addition, indiscriminate use of chemicals in agriculture causes environment pollution and health hazards, destroying the natural balance and beneficial micro-flora of the soil. Moreover, consumers are becoming increasingly concerned about chemical pollution of the environment and pesticide residues on food. Farmers are more often being faced with pathogen's resistance to chemical fungicides. Therefore, it is an urgent need for efficient alternative measures to combat the disease and inoculums buildup.

As alternative of chemical fungicides, biopesticides are now being consided eco-friendly component. In this case, living microorganisms act as antagonist, parasites and predators (Kwok *et al*., 1987). The antagonism of a biological agent reduces pathogen's ability to produce inoculum. In this context, future disease levels will eventually be reduced by such biological control measures (Fokkema, 1995). *Trichoderma* spp. have played a considerable role as biocontrol agent (Papavizas, 1985) and is recognized as an effective biocontrol agent against soil-borne plant pathogenic fungi such as *Fusarium, Sclerotium, Rhizoctonia* etc. (Chet and Inbar, 1994). *Trichoderma* significantly destroys the sclerotia of *S. rolfsii* (Susceelendra and Schlosser, 1999) and it is antagonistic to *S. rolfsii*, overlaps the pathogen and suppresses their growth (Iqbal *et al*., 1995).

Trichoderma produces chemicals called trichodermin which is responsible for its antagonistic properties (Tverdyukov *et al*., 1994). Thus *T. harzianum* may be used as an ecofriendly option to save many beneficial micro-organisms in

the nature. This biocontrol agent would be potential to protect seedlings against diverse soil borne pathogenic fungi. It is also reported that *Trichoderma* has promising contribution in plant growth (Burr *et al*., 1978 and Baker, 1988).

The major limitation of biological control by *Trichoderma* sp. is the production of inoculam in large scale. Many researchers have worked on mass production of *Trichoderma* inocula in the form of spore or other propagules (Papavizas and Lewis 1985; Kenny and Couch 1981 and Churchill 1982). The viable inocula must be produced in an inexpensive medium and the cost of production for treatment of large areas must be competitive with that of the chemical pesticides. The economic mass production of antagonists could be achieved by using readily available crude agricultural product. Various substrates like grain bran (Wells *et al*., 1972), celatom and molasses (Backman and Rodriguez-Kabana, 1975), wheat straw (Akhtar, 1977), wheat bran (Hader *et al*., 1979), cereal meal and sand (Mangenot and Diem, 1979), barley grain (Moity Shatla, 1981),wheat bran and saw dust (Lewis and Papavizas, 1980), sand and corn meal (Lewis and Papavizas, 1980) and combination of different substrates have been used to produce inocula of *Trichoderma* spp (Dubey and Patel, 2002). *Trichoderma* spp need some incubation period for their establishment in soil to act as an antagonist. Considering the above facts the present study was undertaken to achieve the following objectives:

1. To isolate *Trichoderma harzianum* from rhizosphere soil ;
2. To formulate *Trichoderma* based biopesticide using different grain brans and peat soil based substrates;
3. To find out the efficacy of biopesticide against damping off of eggplant and tomato seedlings; and
4. To determine the sustainability of *Trichoderma* in different formulations.

REVIEW OF LITERATURE

Eggplant and tomato are important vegetable in Bangladesh. These seedlings are frequently attacked by damping off pathogen in seed bed. A number of soil born organisms like *Sclerotium rolfsii, Fusarium oxysporum, Pythium* spp. and *Rhizoctonia solani* are involved to cause this disease. This disease is great threat for production of eggplant and tomato in our country. Evidences of research work regarding management of damping off of eggplant and tomato are very limited. However, some available and important findings on various aspects for management of damping off of seedlings has been compiled and presented in this chapter:

Kumar (2013) reported that, *Trichoderma* is a genus of asexually reproducing fungi that is present in all types of soils. Recent reports show that they are opportunistic, avirulent plant symbionts, as well as being parasites on other fungi. A number of successful biocontrol products based on different species of Trichoderma have been commercialized in India, USA and elsewhere in the world.

Kashem *et al*. (2011) conduct a series of experiments to assess the effect of 14 isolates of *Trichoderma* spp. (*Trichoderma harzianum* and *T. viride*) for controlling foot and root rot of lentil caused by *Fusarium oxysporum*. The pathogenecity of 12 isolates of *F. oxysporum* and the mass production of an isolate of *T. harzianum* on 25 substrates are also studied. *Trichoderma* isolates inhibited the growth of *F. oxysporum* up to 92.07 % on agar plates.

Pandya *et* al. (2011) reported that, soil-borne pathogens in fungi cause important losses, being the most aggressive. The distribution of several

phytopathogenic fungi, such as *Pythium*, *Phytophthora*, *Botrytis*, *Rhizoctonia* and *Fusarium* have widely spreaded during the last few years due to change of intensive farming crops culture and environment. *Trichoderma* as a biocontrol agent (BCAs) is well recognized due to their high reproductive capability and show strong aggressiveness against phytopathogenic fungi and efficiency in promoting plant growth and defense mechanisms.

Amin *et al.* (2010) carried out a study on six isolates of *Trichoderma* spp. for their ability to inhibit soil borne pathogens of different vegetables viz., *Rhizoctonia solani* (isolates from tomato), *Sclerotium rolfsii* (causing collar rot of tomato) and *Sclerotinia sclerotiorum* under *in vitro* conditions. Dual culture of pathogens and *Trichoderma* spp. revealed that *T. viride* highly inhibited (65.71%) mycelial growth of *Rhizoctonia solani* over control. In case of *Sclerotium rolfsii* and *Sclerotinia sclerotiorum, T. viride* proved to be potential inhibiting mycelial growth of thet pathogens.

Hossain and Hossain (2010) formulate a *Trichoderma* based BAU-bio fungicide that was found very much effective agent several tikka disease of groundnut, foot and root rot of pulses and diseases of some vegetable crops. BAU-bio fungicide also helpful to control seed borne mycoflora, increasing seed germination and seedling vigour of some vegetables. Management of seedling diseases are successfully possible by using BAU-Biofungicide, biofertilizer and cowdung in blackgram, mungbean and lentil.

Tran (2010) conducted surveys on food crops, industrial crops, vegetable crops and fruit crops in the north and south of Vietnam and reported that *Trichoderma* can be isolated easily from soil, root and plant organic matters. *Trichoderma viride, T. harzianum, T. hamatum* were predominant species in Vietnam. Laboratory and field trials proved that *Trichoderma* species had ability to suppress growth of fungal plant pathogens and enhance plant growth and development. He reported that, *Trichoderma* products have been commercially developed by several companies, institutes and universities such

as: BIMA, Trico-ĐHCT, Promot Plus WP, Vi DK, NLU-Tri, Bio – Humaxin and are available in markets. *Trichoderma* product can be used in many ways including: seed treatment, applied direct to the soil before planting and added to organic fertilizers.

Meah (2007) tested the pathogenecity of 10 isolates of *Sclerotium rolfsii* on eggplant (var. Dohazari) and he found that all the isolates of *S. rolfsii* significantly affected the seed germination, pre-emergence death, damping off, foot rot and plant stand.

Chandrasehar *et al.* (2005) conducted lab and green house experiments to determine the antagonistic effect of *Trichoderma harzianum* against *S. rolfsii* that caused tomato collar rot. They found that *Trichoderma harzianum* in *in vitro* condition completely suppressed the growth of *S. rolfsii* and in green house condition in pot culture increased the percent survival of treated seedling applied as seed treatment and soil drenching.

Islam (2005) also reports while working on controlling of seedling diseases of eggplant that *Trichoderma harzianum* T_{22} effectively controlled damping off disease of seedlings.

Kashem (2005) used soil infestation method for inoculation of *Sclerotium rolfsii*. He found that soil infestation with grain culture at the rate of 0.1% weight basis of dry soil before sowing seeds caused heavy infestation.

Meah and Islam (2005) also found that *Trichoderma* based IPM bio fungicide can effectively control Phomopsis fruit rot, foot/collar and root rot of eggplant and wilt of some vegetables.

Meah *et al*. (2004) reported that *Trichoderma harzianum* cp and *Trichoderma harzianum* T_{22} grown on peat soil based black gram bran was found effective in controlling nursery diseases like damping off, tip over and seedling blight of eggplant and promoted seed germination.

Meah (2003) listed a number of diseases of eggplant caused by fungi, bacteria, virus, nematode and mycoplasma. Of them, collar rot caused by *Sclerotium rolfsii* is damaging to the crop.

Shamsuzzaman *et al*. (2003a) studied for mass production of *Trichoderma harzianum*. Of them, rice straw chick pea bran, rice course with 3% chick pea powder, rice straw with 5% sucrose black gram bran, grass pea bran and peat based wheat bran supported best in mass production of conidia (42.93×10^7/g culture).

Shamsuzzaman *et al*. (2003b) further reported that seed treatment with *Trichoderma harzianum* grown on black gram resulted up to 16.66% higher seed germination , 266.33% fresh shoot weight, 157.14% fresh root weight and 98.55 vigor index of cucurbits over control.

Howlader (2003) reported that *Trichoderma harzianum* cp yielded good result against phomopsis blight and foot rot of eggplant in the field.

Islam *et al*. (2002) evaluated nine organic substrates for their suitability for mass culture of an isolate (GT-1) of *Trichoderma harzianum*, a potential biocontrol agent. They found that maize meal was the best substrate for maximum spore production also colony diameter, mycelial growth was fast compared to others.

Sultana *et al*. (2001) observed growth and storability of *Trichoderma harzianum* and its effect on germination of egg plant seeds. They found that *Trichoderma* treated seed resulted up to 48.62% germination than that of control (untreated).

Chowdhury *et al.* (2000) reported that seed treatment with *Trichoderma harzianum* and *Gliocladium viride* against *Sclerotium rolfsii* resulted up to 21.61% and 48.43% increase in germination in mungbean, black gram, pigeon pea and tomato, respectively and showed good effect on seed born mycoflora. Moreover, significant growth enhancements of mungbean, blackgram and

tomato have been achieved by treating seeds with antagonists. The antagonists were found effective against *Sclerotium rolfsii*.

Das *et al.* (2000) screened five media (wheat bran, maize meal, sand medium, potato dextrose agar and saw dust) for mass multiplication of *Trichoderma koningii* in vitro. Wheat bran proved to be more promising for the growth and sporulation of the fungi. Growth and sporulation of *Trichoderma* spp. were significantly highest after 14 days, than 7 days of inoculation.

Rettinassababady and Ramadoss (2000) reported that *Trichoderma* spp. were mass multiplied in black ash, coir waste, farmyard manure, rice husk, spent straw from mushroom bed, sugarcane bagasse, talc and vermiculite. *Trichoderma* growth and spore production was maximum in farmyard manure and coir waste (474×10^5, 263×10^5 spore/g) in 3 weeks in culture.

Sultana and Hossain (1999) evaluated *Trichoderma harzianum* for controlling foot and root rot (*Fusarium oxysporum* and *Sclerotium rolfsii*) of Lentil cv. BARI Masur-1 under field condition. Seeds of lentil treated with *Trichoderma harzianum* (2×10^6 conidia/seed) contributed 47.85% to 112.49% reduced of foot and root rot diseased plants over control. *Trichoderma harzianum* treated seeds increased germination up to 13.37% and resulted up to 3.69% more field emergence over control. *Trichoderma harzianum* treated seeds resulted yield up to 1783.33 kg/ha that accounted 81.60% higher seed yield.

Shamarao *et al*. (1998) tried mass multiplication and sporulation of *Trichoderma viride* using different substrates like oil cake, farmyard manure, wheat bran, poultry manure, dung, jaggery, groundnut cake, neem cake and pongamia. Wheat bran was the most suitable substrate for sporulation of the antagonists.

Begum (1997) selected four *Trichoderma* spp. and evaluated their antagonistic potential against the major soil-borne plant pathogens *Sclerotium rolfsii, Fusarium oxysporum* and *Macrophomina phaseolina.* Two induced mutants of

Trichoderma spp. showed better performances than control strain in reducing the seedling mortality in chickpea and lentil caused by *Fusarium oxysporum* and *Sclerotium rolfsii* under glasshouse condition

Das *et al.* (1997) screened five media (wheat bran, rice bran, maize meal, sand medium, potato dextrose agar and saw dust) for mass multiplication of *Trichoderma viride*, *T. harzianum* and *T. koningii in vitro*. Wheat bran proved to be more promising for the growth and sporulation of the fumgi. Growth and sporulation of *Trichoderma* spp. were signifantly higher after 14 days, than after 7 days of inoculation.

Roberti *et al*. (1996) investigated the activity of *Trichoderma harzianum* 74 on bean (*Phaseolus vulgaris*) rot caused by *Scelorotium rolfsii* when applied to seeds. *Trichoderma* strains were active in bean root rot ensuring control of *Scelorotium rolfsii*. *Trichoderma harzianum* reduced the growth of *Scelorotium rolfsii* and parasitized *Scelorotium rolfsii*, hyphae by direct contact, forming coils, short contact branches and hook-shaped hyphal tips.

Mukherjee *et al.* (1995) observed that *Trichoderma harzianum* was effective in suppressing *Sclerotium rolfsii* and *Rhizoctonia solani*. *Trichoderma harzianum* was found to be effective in destroying the sclerotic of both fungi.

Mukhopadhyay (1995) stated that two bio-agents viz., *Gliocladium virens* and *Trichoderma harzianum* were used for treating seeds of various crops, like chickpea, lentil, groundnut, tomato and cauliflower for protection against wide range of soil brone pathogens viz., *Rhizoctonia solani*, *Sclerotium rolfsii*, *Pythium spp.* and *Fusarium oxysporum*. Such biological treatment was also integrated with suitable fungicide in view of the insensitivity of the bio-agents to some chemicals. The treatment was found highly effective and resulted in enhanced crop performance when compared with biological or chemical treatment alone.

Inber *et al*. (1994) *Trichoderma harzianum* to cucumber seedlings as a peat-bran preparation incorporated into the propagation mixture in a commercial

plant production nursery. Increase of 23.8% in seedlings height and 96.1% in leaf area were recorded. On marketing day (after 18 and 30 d) recorded significant DW compared with untreated control plants. *Trichoderma*-treated seedlings were more developed, grew more vigorously and contained higher levels of cholorophyll than control plants. No significant differences were found in N, P or K content between treatments. Cucumber seedlings which were transplanted to a commercial greenhouse were analyzed over 2 successive growth cycles following soil fumigation with methyl bromide (500 kg/ha). Results revealed that the *Trichoderma* treated plants were more resistant to damping off diseases caused by *Pythium* spp. and *Rizoctnia solani*. During the first cycle, immediately after soil fumigation, no damping-off observed with either treatment, except in border beds where 4% of the non treated plants died, compared with no damping-off in the *Trichoderma* treated plants. However, significant reductions in damping-off by 67% and 52% were obtained in middle and border, respectively, during the 2^{nd} growing cycle compared with untreated controls.

Chet and Inbar (1994) studied on biological control of fungal pathogens and reported that *T. harzianum* as effective bio-control agent of soil-borne plant pathogenic fungi. Lectins were found to be involved in the recognition between *Trichoderma* spp. and its host fungi, where as Chitinase is involved in the degradation of the host wall.

Sugha *et al*. (1993) reported that conidial coating of the antagonistic *Trichoderma harzianum* and *T. viride* on seeds significantly reduce seedling mortality (47-65%) infected by *Sclerotium rolfsii* compared with untreated controls.

Sangeetha *et al*. (1993) found farmyard manures as the best for formulation of *Trichoderma viride* and *Trichoderma harzianum* followed by wheat bran and rice bran. Peat soil alone and rice straw were found as poor substrates.

Xu *et al.* (1993) observed that both isolates of *Trichoderma* T82 and NF9 inhibited hyphal growth of *Sclerotium rolfsii, Rhizoctonia solani, Pythium aphanidermatum, P. spinosum* and *Fusarium oxysporum.* In greenhouse experiments, soil treatment with 0.6 % (w/w) T82 bran culture (10^7 CFU /g) reduced incidence of disease caused by *S. rolfsii, R. solani* and *P. aphanidermatum* by 46.5%, 28.4% and 81.2% respectively, 20 days after inoculation with the pathogens. Seed treatment with T82 or NF9 spore suspension (10^8 CFU /ml) increased emergence of cucumber seedlings by 14% and 20%, respectively, 11 days after inoculation with *S. rolfsii.*

Kaur and Mukhapadhyay (1992) reported that integrated use of *Trichoderma harzianum* with fungicidal seed treatments in the fields significantly reduced the incidence of chickpea wilt complex and increased crop yield. Seed treatment with vitavax-200 (Carboxin + Thiram) and Ziram resulted 29.9% disease control. This control increased to 63.3% when *Trichoderma harzianum* was added.

Monaco *et al*. (1991) used for treating seeds as bio-control agents of *Fusarium* and *Sclerotium.* They isolated *Trichoderma harzianum, Trichoderma koningii* and *Trichoderma aureoviride* from tomato fields in the horticultural area of Laplanta, Argentina, naturally infected with *Fusarium* spp. and *Sclerotium (Corticum) rolfsii*. All 3 species of *Trichoderma* were effective against *Fusarium* spp. and *Corticium rolfsii in vitro* and in subsequent field trials. Seedling emergence was significantly increased when *Trichoderma harzianum* were applied to seeds sown in soil infected with the pathogens. They also reported that each *Trichoderma* spp. was effective against *C. rolfsii*.

Haque *et al*. (1990) used *Trichoderma harzianum* as biocontrol agent for controlling root rot diseases of okra, sunflower, soybean and mungbean. *Trichoderma harzianum* used as seed treatments or as soil drenches for the control of root rot caused by *Macrophomina phaseolina, Rhizoctonia solani* of

sunflower, soybean and *Vigna radiate* under field conditions. *Trichoderma* showed excellent inhibitory effect of controlling *Fusarium* and *Rhizoctonia*.

Kumar and Khare (1990) found *Trichoderma harzianum* as antagonistic to *Sclerotium rolfsii* when soybean seed were treated with *Trichoderma harzianum, Gliocladium virens, Bacillus subtilis* and *Streptomyces* spp. They also showed that *Fusarium* infection of sunflower was reduced by *Trichoderma harzianum.*

Krishnamoorthy and Bhoskaran (1990) reported that soil inoculation with *Trichoderma harzianum* and *Trichoderma viride* gave good control of *Sclerotium rolfsii* and in treated pots gave 78.2% and 72.2% egg plant seed germination respectively compared to 19.3% in the control.

Ordentlich and Chet (1989) conducted an experiment in greenhouse and found that *Trichoderma harzianum* obtained from field soil were effective for control diseases (caused by *Sclerotium [Corticium] rolfsii*, *Rhizoctonia solani*, *Pythium*, *Fusarium* of various crops) when grown in a semi-solid fermentation medium on wheat bran: peat.

Harman *et al.* (1989) reported on combining effective strains of *Trichoderma harzianum* and solid matrix priming to improve biological seed treatments. They developed progeny strains (T_{12} and T_{95}) by fusing two strains of *Trichoderma harzianum* and two of which were selected for further study. Seeds of cotton, cucumber, pea, snap bean, maize and wheat were also planted in soil infested with *Pythium ultimum* and *Rhizoctonia solani*. In all crop pathogen combinations, seed treatments with parental and progeny *Trichoderma strs* with or without solid matrix priming increased stands relative to the untreated control and were as effective as vitavax-200 (Carboxin + Thiram).

Shin *et al.* (1987) found that soil treated with *T. viride* reduces damping off of sesame seedlings. The sesame seedlings on beds treated with the antagonist grew better than seedlings in untreated soil. Soil and seed treated with *T. viride*

reduced sunflowers infection (*S. sclerotiorum* and *B. cinerea*) in the glasshouse and prevented infection also in the field.

Jabos and Kamoen (1986) found that *Trichoderma harzianum* produced cell wall lysine enzymes which developed antagonism against plant pathogens and improved biological control.

Sivan and Chet (1986) prepared wheat bran / peat mixture (1:1v/v) adjusted to 40% moisture (w/w) autoclaved for 1hr at 121^0C. The substrate mixture was inoculated with a conidial suspension of *Trichoderma* and incubated in an illuminated chamber at 30^0C. This preparation of *Trichoderma* was mixed with soil (5g/kg soil) before sowing seeds of the test plants.

Waraitch *et al*. (1986) used soil mixing method of inoculation of *Sclerotium rolfsii* (multiplication on sterilization sorghum seeds pre-soaked in 2% sucrose solution) was mixed in soil near the plants @ three 500 ml flask per 100 m^2.

Strashnow *et al.* (1985) reported that application of *T. harzianum* to soil or by coating tomato fruits was found to reduce *R. solani* fruit rot by up to 43% and 85% respectively under laboratory conditions. When it was mixed with naturally infested soil. *T. harzianum* reduced the *R. solani* inoculum potential of soil by 86% and fruit rot by 27-51%).

Sivan *et al*. (1984) used wheat bran/peat preparation of *Trichoderma harzianum* mixed with lomy sand (5g/kg soil) artificially infested with *Pythium aphanidermatum* significantly reduced disease incidence caused by this pathogen in cucumber, pea and tomato at 69,81 and 85% respectively.

Mirkova (1982) studied the antagonistic activity of *Trichoderma* spp. against some pathogens and reported that among 5 *Trichoderma* spp, 3 isolates of *Trichoderma harzianum* were most antagonistic.

Elad *et al*. (1982) studied on the prevention of plant infection by biological means. *Trichoderma harzianum* isolated from the soil showed on antibiotic activity against *Sclerotium rolfsii* when grown on cell walls of the pathogens. It

produced extra cellular B (1-3) glucose and chitinase when applied in the form of wheat bran culture to soil infested with *Sclerotium rolfsii* in the glass house. *Trichoderma harzianum* effectively controlled damping off of eggplant.

Agrawal *et al*. (1977) found filtrates of *Trichoderma* inhibited the growth of *Sclerotium rolfsii* on PDA, in pot trial the antagonist controlled seedling death. Culture was more effective when applied to seed rather than soil.

Wells *et al*. (1972) found *Trichoderma harzianum* pathogenic to *Sclerotium rolfsii* in agar medium. They reported *Trichoderma harzianum* to effectively control *Sclerotium rolfsii* on peanuts, tomatoes and blue lupins under greenhouse condition and also under greenhouse condition and also under field condition when applied (1-3 times) over the plants on to the soil surfaces.

METHODOLOGY MATERIALS AND METHODS

3.1 Experimental Site

The experiments were conducted at the central laboratory as well as in the nursery house Department of Plant Pathology, Sher-e-Bangla Agricultural University, Dhaka, 1207. Plastic tray or soil pots were used as unit plot.

3.2 Experimental Period

The experiments were carried out during the period from December 2013 to September 2014.

3.3 Isolation of *Trichoderma*

Trichoderma sp. was isolated from rhizosphere soil from four different Districts (Gazipur, Dhaka, Comilla and Rangpur) in Bangladesh by soil dilution technique.

Trichoderma spp. was isolated by dilution plate technique (Dhingra and Sinclair, 1985) as described below:

v. Disinfection of working area

Since the bacteria and fungi are always present as contaminants in the soil, it is important to exclude them as much as possible from the surface of the working area and the equipment to be used. The surface of the working area was disinfected with cotton soaked in methylated spirit (70%). The hands and equipments were disinfected by the same means. The glass wares (test tubes, petri dishes, pipettes, beakers etc.) were sterilized in dry oven.

vi. Preparation of working samples

For every soil samples, working sample was prepared from the composite sample that was made in immediate after collection of

sample from rhizosphere root at flowering stage of five important vegetable crops.

vii. Making suspension (soil dilution)

a. 1gm of the soil was taken in test tube containing 9 ml of sterile water and stirred thoroughly for few minutes in order to obtain an uniform 1:10 soil suspension. This solution was used as stock suspension.

b. 1ml of that 1:10 stock suspension was transferred with the help of sterile pipette into the 2nd test tube containing 9 ml sterile water and shaken thoroughly to make 10^{-1} dilution.

c. 1ml of the 10^{-1} dilution is transferred to 3rd test tube containing 9 ml sterile water by sterile pipette to make 10^{-2} dilution. In this way dilution was made up to 10^{-4}

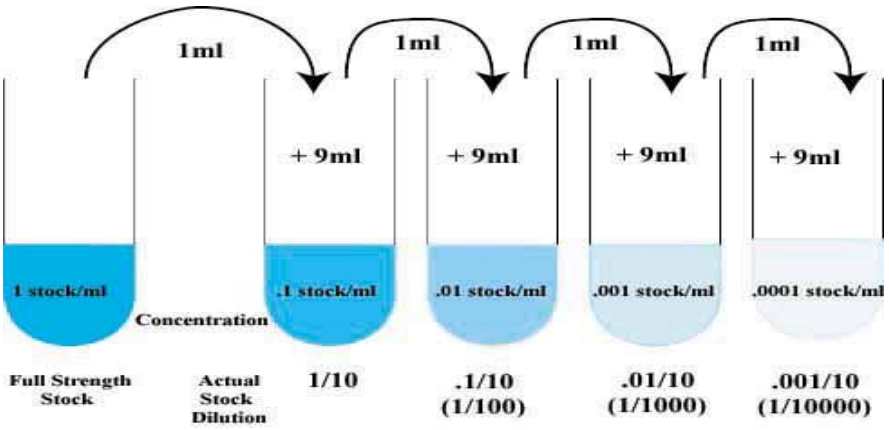

Figure 1. Preparation of dilution series of soil sample

viii. Isolation of micro-organisms (*Trichoderma* sp.) from soil

a. 20 ml of warm (approx. 45°C) melted PDA medium was poured in each sterile petri-plate.

b. 1 ml of diluted soil sample (10^{-4}) was placed at the center of PDA and spreaded. Four petri-dishes were inoculated with 1 ml of each diluted sample.

c. The inoculated PDA plates were incubated for 7-10 days at room temperature (25±1^{o}C).

d. The colonies grown out on PDA were recorded after 3-5 days of incubation. Sub cultures were made by transferring a small colony to a new petri-dish on the basis of color and morphology of the colony. Further recultures were made for purification. The contaminated plates were discarded.

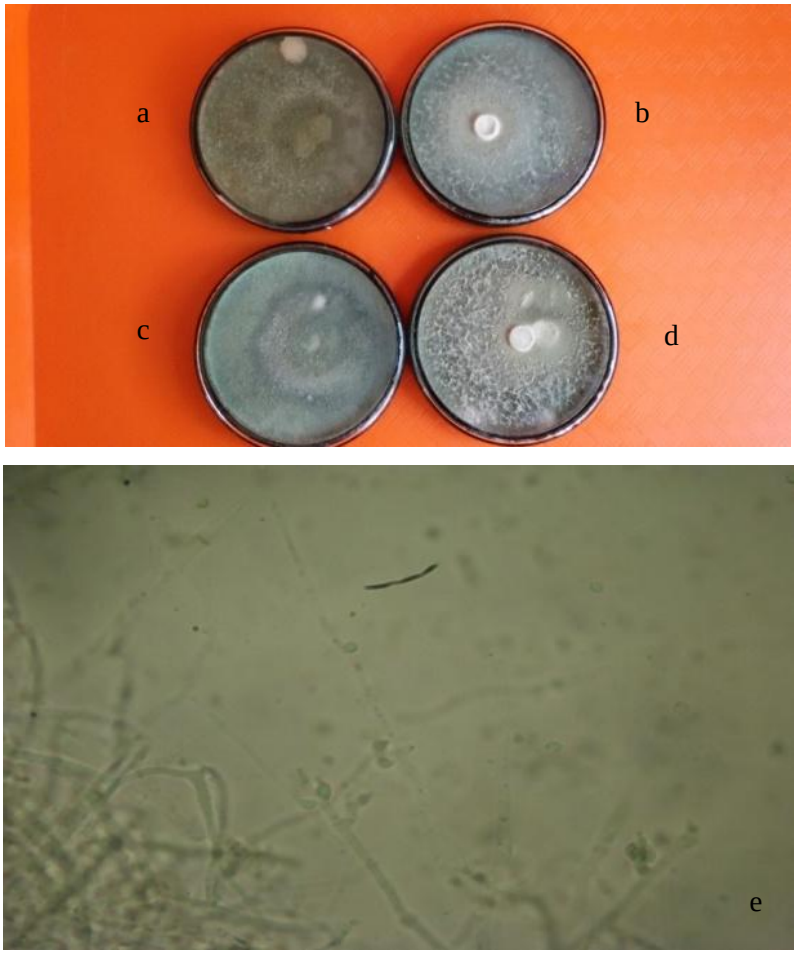

Plate 1. Isolates of *Trichoderma harzianum* (a) TJG – *Trichoderma harzianum*, Joydevpur, Gazipur (b) TSD - *Trichoderma harzianum*, Sher-e-Bangla Nagar, Dhaka (c) TCC - *Trichoderma harzianum*, Chandina, Comilla (d) TTR - *Trichoderma harzianum*, Taragonj, Rangpur (e) Conidia and mycelium of *Trichoderma harzianum*

3.4 Collection of peat soil

Peat soil was collected from Tungipara, Gopalgonj, Bangladesh.

3.5 Collection of substrates

Substrates were collected from local shops of Kawran Bazar, Dhaka. Substrates were kept in 4^0C until use.

3.6 Collection of seeds

Eggplant and tomato seeds were collected from Bangladesh Agricultural Development Corporation (BADC), Gabtoli, Dhaka.

3.7 Variety used

1. Eggplant – BARI Brinjal-1 (Uttara)
2. Tomato – BARI Tomato-2 (Raton)

3.8 Treatments of the experiment

All together 8 peat soil based substrates along with a control were explored in the experiment stated below:

1. Rice bran + Peat soil + Water (1:1:2)
2. Wheat bran + Peat soil + Water (1:1:2)
3. Lentil bran + Peat soil + Water (1:1:2)
4. Gram bran + Peat soil + Water (1:1:2)
5. Black gram bran + Peat soil + Water (1:1:2)
6. Mustard oil cake + Peat soil+ Water (1:1:2)
7. Grass Pea bran + Peat soil+ Water (1:1:2)
8. Saw Dust + Peat soil+ Water (1:1:2)
9. Control

3.9 Sterilization of Substrates and Inoculation of *Trichoderma* in the multiplication

The requisite amount of materials for each substrate was thoroughly mixed in a 1000 ml Erlenmeyer flask and autoclaved at 121^0C for 15 minutes for

sterilization. The sterilized substrates allowed to cool down and then inoculated with 5 mm dia mycelia disc of 7 days old *Trichoderma* culture. Seven discs for each flask were used for inoculation. Inoculated flasks were then incubated at room temperature $(25\pm2)^{0}$C.

a

b

Plate 2. Mass multiplication of *Trichoderma* in different substrates (a) sterilized substrates before inoculation (b) Sterilized substrates after inoculation

3.10 Formulation and measurement of spore/g substrate

After incubation for 25 days, the contents were taken out from the flasks, air dried in laminar airflow cabinet and grinded in a blender. The grinded materials were kept in polythene bag with labeling and treated as formulated *Trichoderma*. The spores of *Trichoderma* per gram of formulated products were measured by Haemocytometer.The number of conidia per plate was determined with the help of Haemocytometer following the procedure of Ashrafuzzaman (1976).

Plate 3. Formulated *Trichoderma* in polythene bag

3.11 Collection/isolation and Maintenance of *Sclerotium rolfsii*

The pathogen was obtained from naturally infected tomato plant grown in the experimental field of the Department of Plant Pathology, SAU, Dhaka. The typical collar rot symptoms of tomato plant showed a rot with dry black to brown black lesions around the stem at collar region. The plant was still alive with pale green, and reduced sized leaves. Numbers of round brown to black sclerotia were found.

The infected tissue of the collar region of the plant was collected and repeatedly washed in fresh water and surface was sterilized with 10% Clorox for 1 minute followed by three times washing in distilled water.

Then the pieces of infected tissue were placed on PDA acidified with one drop of 5% lactic acid and inoculated at 22± 2ºC for 7days. After incubation, white mycelia and sclerotia were formed (Plate 4). The pathogen was purified and multiplied subsequently through hyphal tip culture on PDA, for preparation of inocula.

Plate 4. Culture of *Sclerotium rolfsii* (7 days old)

3.12 Preparation of inocula of *Sclerotium rolfsii*

Barley culture method was followed to culture and multiply *Sclerotium rolfsii*. Inocula of *Sclerotium rolfsii* were prepared in barley culture. Barley grains collected from market were throughly washed in water and kept soaked in fresh water for 24hrs. After decantation, barley grains were taken in 500 ml Erlenmeyer flask at the rate of 200g in each. The flasks were plugged with cotton followed by wrapping the mouth with brown paper. The flasks containing moist barley grains were sterilized in autoclave at 121ºC under 15*lbs* pressure for 15 minutes. The sterilized barley grains in the flask were cooled and inoculated aseptically with mycelial blocks (5mm) of pure culture of *Sclerotium rolfsii* on PDA and inoculated at room temperature for 7-8 days. The flasks were shaken periodically with hand for proper distribution of fungal mycelium throughout the entire mass of the inoculated barley grains (Plate 6).

The mycelial growth of the fungus covered entire barley mass in the flask when small round white sclerotia started to form. It was taken out of the flask after fifteen days. The entire mass was spread on brown paper and air dried at room temperature. The colonized dried barley grains were used as inocula for inoculation of plants (Plate 7) (Babar, 1999).

Plate 5. Air drying of barley culture of *Sclerotium rolfsii*

a **b**

Plate 6. Barley culture of *Sclerotium rolfsii* (a) After inoculation of 5 days (b) After inoculation of 15 days

3.13 Preparation of soil

Soil and cow dung were mixed in (2:1) ratio and kept for 15 days and then the soil sterilized with 5 ml formalin (40%) diluted with 20 ml water for 4 kg soil (Dasgupta, 1988) and the prepared soil was heaped in square block. Soil heap was covered by polyethylene sheet for 48 hr. After 4 days of treatment plastic tray and pot were filled up with the sterilized soil.

3.14 Application of formulated *Trichoderma harzianum* and *Sclerotium rolfsii* in the soil

Formulated *Trichoderma* were mixed with the soil of each plastic trays and earthen pots (except control) @ of 20g/kg soil. The treated soil was incubated for 7 days maintaining proper soil moisture. After that, soil was inoculated with barley grain colonized by *Sclerotium rolfsii* @ 20g/kg of soil. Inoculated soil was incubated for 7 days maintaining proper soil moisture.

Plate 7. Application of formulated *Trichoderma harzianum* in plastic tray and earthen pot

Plate 8. Application of barley culture of *Sclerotium rolfsii*

3.15 Sowing seeds in trays and earthen pots

One eighty seeds of tomato plant were sown in each tray and one hundred seeds of eggplant were sown in each earthen pot after 7 days of application of formulated *Trichoderma* and *Sclerotium rolfsii*.

3.16 Data collection

Data was collected on following parameters-

i. % of seed germination
ii. % pre-emergence damping off
iii. % post-emergence damping off
iv. % Tip over
v. Seedling height (cm)
vi. Vigor index
vii. Fresh weight of seedling (gm)

3.17 Statistical analysis of data

The collected data obtained for various parameters were coded, tabulated and analyzed by MSTAT-C computer package programme.

Plate 9. A view of eggplant seedlings in the pots under investigation

Plate 10. A view of tomato seedlings in the trays under investigation

RESULTS

The results obtained from the present study on the effect of nine different treatments viz. T_1 (*Trichoderma* + Rice bran + Peat soil + Water), T_2 (*Trichoderma* + Wheat bran + Peat soil + Water), T_3 (*Trichoderma* + Lentil bran + Peat soil + Water), T_4 (*Trichoderma* + Gram bran + Peat soil + Water), T_5 (*Trichoderma* + Black gram bran + Peat soil + Water), T_6 (*Trichoderma* + Mustard oil cake + Peat soil + Water), T_7 (*Trichoderma* + Grass Pea bran + Peat soil + Water), T_8 (*Trichoderma* + Saw Dust + Peat soil + Water), T_9 (Control) for the management of damping off of vegetable seedlings are presented in this chapter.

4.1 Number of spore/mm^2 (×10^4) in different *Trichoderma* isolates

Nubmer of spores in different *Trichoderma harzianum* isolates was presented in Table 1. The highest number of spore/mm^2 was observed in isolate T_1 (TJG- *Trichoderma harzianum*, Joydevpur, Gazipur) (6.42×10^4) followed by isolate T_2 (TSD - *Trichoderma harzianum*, Sher-e-Bangla Nagar, Dhaka) (5.70×10^4). The lowest number of spore/mm^2 was observed in isolate T_4 (TTR - *Trichoderma harzianum*, Taragonj, Rangpur) (5.13×10^4) preceded by isolate T_3 (TCC - *Trichoderma harzianum*, Chandina, Comilla) (5.30×10^4). Based on the higher spore production isolate T_1 (TJG - *Trichoderma harzianum*, Jodevpur, Gazipur) was considered to make the subsequent formulation.

Table 1. Inoculum potential (spore/mm^2) by different *Trichoderma* isolates assayed in the experiment

Trichoderma Isolate	No. of spore/mm^2($\times 10^4$)
T_1 - TJG	6.42 a
T_2 - TSD	5.70 b
T_3 - TCC	5.30 c
T_4 - TTR	5.13 c
$LSD_{(0.05)}$	0.31
CV (%)	2.97

T_1= (TJG - *Trichoderma harzianum*, Joydevpur, Gazipur)

T_2= (TSD - *Trichoderma harzianum*, Sher-e-Bangla Nagar , Dhaka)

T_3= (TCC - *Trichoderma harzianum*, Chandina , Comilla)

T_3= (TTR - *Trichoderma harzianum*, Taragonj, Rangpur)

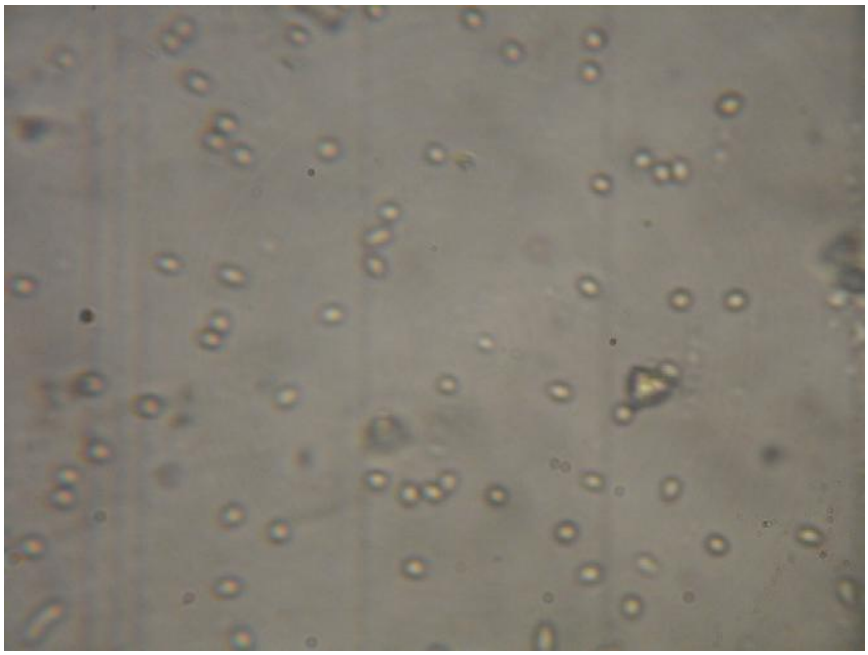

Plate 11. Spores of *Trichoderma* isolate TJG (*Trichoderma harzianum*, Joydevpur, Gazipur) in haemocytometer observed under compound microscope

4.2 Effect of different substrates on mass multiplication of *Trichoderma harzianum*

Inoculum potentials in respect of number of spore of *Trichoderma harzianum* in different treatments was presented in Table 2. All the treatments differed significantly in terms of number of spores of *Trichoderma harzianum*. The highest number of spore/gm (24.27×10^7) was observed in T_1 where black gram bran and peat soil were mixed with water at 1:1:2 ratio followed by T_2 (22.23×10^7) where grass pea bran was mixed with peat soil and water at 1:1:2 ratio. The third highest number of spore/gm (21.23×10^7) was observed in T_3 (Gram bran + Peat soil + Water, ratio -1:1:2) followed by treatment T_4 (Wheat bran + Peat soil + Water, ratio -1:1:2) (20.23×10^7). The lowest number of spore/gm was observed in T_8 (Saw Dust + Peat soil + Water, ratio -1:1:2) (10.23×10^7) preceded by treatment T_7 (16.23×10^7). In case of T_5 and T_6 number of spore/gm were 19.50×10^7 and 18.23×10^7.

Table 2. Effect of different substrates on mass multiplication of *Trichoderma harzianum*

Treatment	No. of spore/gm($\times 10^7$)
T_1	24.27 a
T_2	22.23 b
T_3	21.23 c
T_4	20.23 d
T_5	19.50 e
T_6	18.23 f
T_7	16.23 g
T_8	10.23 h
$LSD_{(0.05)}$	0.49
CV (%)	1.48

T_1= *Trichoderma* + Black gram bran + Peat soil + Water

T_2= *Trichoderma* + Grass pea bran + Peat soil + Water

T_3 = *Trichoderma* + Gram bran + Peat soil + Water

T_4 = *Trichoderma* + Wheat bran + Peat soil + Water

T_5 = *Trichoderma* + Rice bran + Peat soil + Water

T_6 = *Trichoderma* + Lentil bran + Peat soil + Water

T_7 = *Trichoderma* + Mustard oil cake + Peat soil + Water

T_8 = *Trichoderma* + Saw dust + Peat soil + Water

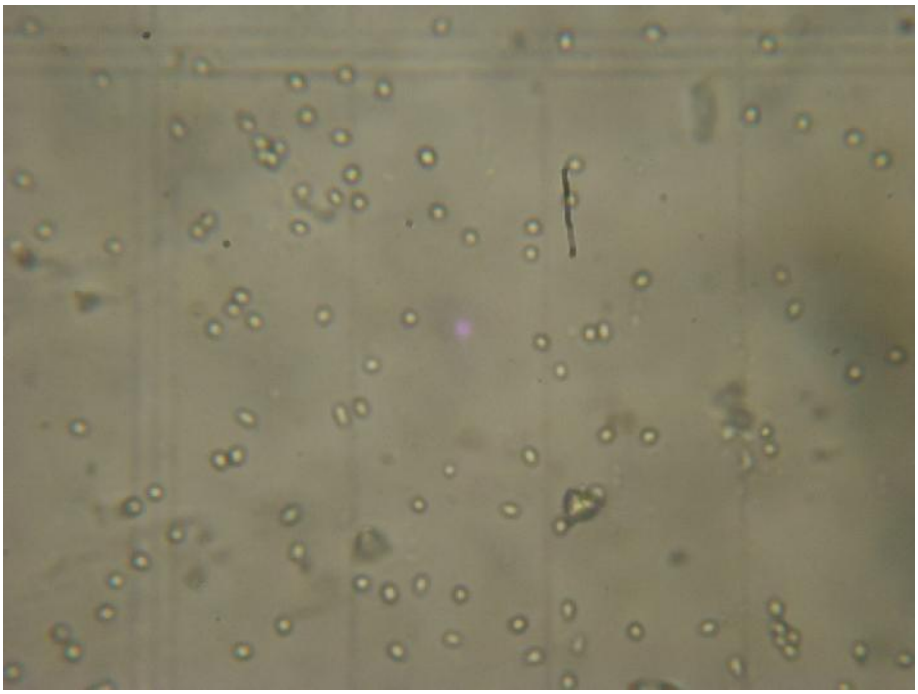

Plate 12. Spores of *Trichoderma* isolate TJG (*Trichoderma harzianum*, Joydevpur, Gazipur) in treatment T_5 (*Trichoderma* + Rice bran + Peat soil + Water) in Haemocytometer observed under compound microscope

4.3 Effect of different treatments on germination of eggplant seedlings at 10 days after sowing

The effect of treatments on germination of eggplant seedlings at different days after sowing (DAS) was presented in Table 3. The effect of different treatments on germination of eggplant seedlings differed significantly in comparison to control. At 10 DAS, the highest germination (76.00%) was observed in T_5 (*Trichoderma* + Black gram + Peat soil + Water) and the lowest germination (44.00%) was observed in T_9 (Control). The second highest germination (71.67%) was observed in T_7 (*Trichoderma* + Grass pea bran + Peat soil + Water). The effect of treatment T_2 (*Trichoderma* + Wheat bran + Peat soil + Water) on germination at 10 DAS was statistically similar with the effect of treatment T_1 (*Trichoderma* + Rice bran + Peat soil + Water). The effect of treatment T_6 (*Trichoderma* + Mustered oil cake + Peat soil + Water) on germination at 10 DAS was statistically similar with the effect of treatment T_8 (*Trichoderma* + Saw dust + Peat soil + Water).

4.4 Effect of different treatments on the germination of eggplant seedlings at 13 days after sowing

A remarkable effect was observed among the treatments on the germination at 13 DAS of eggplant seedling. The effect of different treatments on germination of eggplant seedlings differed significantly in comparison to control. The highest germination (77.00%) was observed in T_5 (*Trichoderma* + Black gram + Peat soil + Water) and the lowest germination (45.33%) was observed in T_9 (Control). The second highest germination (72.67%) was observed in T_7 (*Trichoderma* + Grass pea bran + Peat soil + Water) which was statistically similar with treatment T_4 (*Trichoderma* + Gram bran + Peat soil + Water) (69.00%). The effect of treatment T_2 (*Trichoderma* + Wheat bran + Peat soil + Water) on germination at 13 DAS was statistically similar with the effect of treatment T_1 (*Trichoderma* + Rice bran + Peat soil + Water). The effect of treatment T_6 on germination at 10 DAS was statistically similar with the effect of treatment T_8.

4.5 Effect of different treatments on the germination of eggplant seedlings at 16 days after sowing

The effect of different treatments on germination of eggplant seedlings varied significantly in comparison to control. At 16 DAS, the highest germination (78.00%) was observed in T_5 (*Trichoderma* + Black gram + Peat soil + Water) and the lowest germination (46.33%) was observed in T_9 (Control). The second highest germination (72.67%) at the same days after sowing was observed in T_7 (*Trichoderma* + Grass pea bran + Peat soil + Water) which was statistically similar with treatment T_4 (70.00%). The effect of treatment T_2 (*Trichoderma* + Wheat bran + Peat soil + Water) on germination at 13 DAS was statistically similar with the effect of treatment T_1 (*Trichoderma* + Rice bran + Peat soil + Water). The effect of treatment T_3 on germination at 13 DAS was statistically similar with the effect of treatment T_6. According to the performance of the treatments the highest increase of seed germination (68.36%) was counted in T_5 (*Trichoderma* + Black gram + Peat soil + Water) followed by T_7 (56.85%) (*Trichoderma* + Grass pea bran + Peat soil + Water), T_4 (51.10%) (*Trichoderma* + Gram bran + Peat soil + Water) and T_2 (40.29%) (*Trichoderma* + Wheat bran + Peat soil + Water) respectively.

Table 3. Effect of different treatments on the germination of eggplant seedlings at different days after sowing (DAS)

Treatment	% Germination			% Germination increased over control
	10 DAS	13 DAS	16 DAS	16 DAS
T_1	61.00 d	62.00 cd	63.00 cd	35.98
T_2	63.00 d	64.00 c	65.00 c	40.29
T_3	57.00 e	58.00 de	60.00 de	29.50
T_4	68.00 c	69.00 b	70.00 b	51.10
T_5	76.00 a	77.00 a	78.00 a	68.36
T_6	53.00 f	55.00 ef	57.33 e	23.74
T_7	71.67 b	72.67 b	72.67 b	56.85
T_8	50.00 f	51.00 f	52.00 f	12.24
T_9	44.00 g	45.33 g	46.33 g	-
$LSD_{(0.05)}$	3.09	4.15	4.35	-
CV (%)	2.99	3.93	4.05	-

T_1= *Trichoderma* + Rice bran + Peat soil + Water

T_2= *Trichoderma* + Wheat bran + Peat soil + Water

T_3 = *Trichoderma* + Lentil bran + Peat soil + Water

T_4 = *Trichoderma* + Gram bran + Peat soil + Water

T_5 = *Trichoderma* + Black gram bran + Peat soil + Water

T_6 = *Trichoderma* + Mustard oil cake + Peat soil + Water

T_7 = *Trichoderma* + Grass pea bran + Peat soil + Water

T_8 = *Trichoderma* + Saw dust + Peat soil + Water

T_9= Control

a

b

Plate 13. Healthy seedlings of eggplant raised on treatment T_5 (*Trichoderma* + Black gram bran + Peat soil + Water (a) 10 days after sowing (b) 16 days after sowing.

4.6 Effect of different treatments on the post-emergence damping off of eggplant seedlings at 10 days after sowing

The effect of different treatments varied significantly in respect of post-emergence damping off percentage at 10 DAS (Table 4). The highest percent post-emergence damping off was recorded in control (25.00%) and the second highest post-emergence damping off at 10 DAS was observed in treatment T_8 (*Trichoderma* + Saw dust + Peat soil + Water) which was statistically similar with T_3 (*Trichoderma* + Lentil bran + Peat soil + Water). The effect of treatment T_2 on post-emergence damping off at 10 DAS was statistically similar with treatment T_4. The effect of treatment T_7 (*Trichoderma* + Grass pea bran + Peat soil + Water) on post-emergence damping off at 10 DAS was 7.00%. The lowest percentage of post-emergence damping off (4.00%) was recorded in treatment T_5 (*Trichoderma* + Black gram + Peat soil + Water).

4.7 Effect of different treatments on the post-emergence damping off of eggplant seedlings at 13 days after sowing

A remarkable effect was observed among the treatments in controlling post-emergence damping off disease of eggplant at 13 days after sowing (Table 4). The treatments effects differed significantly in terms of post-emergence damping off percentage. The highest effect against post-emergence damping off (6.00%) was observed in case of treatment T_5 (*Trichoderma* + Black gram + Peat soil + Water). The second lowest percent post-emergence damping off (10.00%) was observed in treatment T_7 (*Trichoderma* + Grass Pea bran + Peat soil + Water) which was statistically similar with treatment T_4. The effect of treatment T_8 (*Trichoderma* + Saw dust + Peat soil + Water) on post-emergence damping off at 13 DAS was statistically similar with the treatment T_6 and T_3. The highest percent of post-emergence damping off (28.00%) was observed in case of treatment T_9 (Control).

4.8 Effect of different treatments on the post-emergence damping off of eggplant seedlings at 16 days after sowing

The effect of the treatments on post-emergence damping off percentage of eggplant seedlings at 16 days after sowing (DAS) was presented in Table 4. All the treatments differed significantly in terms of post-emergence damping off percentage. The highest percent post-emergence damping off was counted in control (30.00%) followed by treatment T_8 (23.67%). The effect of treatment T_6 on post-emergence damping off at 16 DAS was statistically similar with the treatment T_3 and T_1. The lowest percent post-emergence (6.33%) was observed in case of treatment T_5 (*Trichoderma* + Black gram + Peat soil + Water). According to the performance of the treatments the highest reduction of post-emergence damping off (78.90%) was counted in T_5 (*Trichoderma* + Black gram + Peat soil + Water) followed by T_7 (65.57%) (*Trichoderma* + Grass pea bran + Peat soil + Water), T_4 (57.77%) (*Trichoderma* + Gram bran + Peat soil + Water) and T_2 (43.33%) (*Trichoderma* + Wheat bran + Peat soil + Water) respectively.

Table 4. Effect of different treatments on post-emergence damping off of eggplant seedlings at different days after sowing (DAS)

Treatment	% Post-emergence damping off			% post-emergence damping off reduced over control
	10 DAS	13 DAS	16 DAS	16 DAS
T_1	15.00 d	17.00 cd	19.33 c	35.57
T_2	12.33 e	15.00 d	17.00 d	43.33
T_3	17.67 bc	19.00 bc	19.67 c	34.43
T_4	11.00 e	11.33 e	12.67 e	57.77
T_5	4.000 g	6.00 f	6.33 g	78.90
T_6	17.00 c	20.00 b	20.00 c	33.33
T_7	7.00 f	10.00 e	10.33 f	65.57
T_8	19.33 b	21.00 b	23.67 b	21.10
T_9	25.00 a	28.00 a	30.00 a	-
$LSD_{(0.05)}$	1.89	2.24	1.71	-
CV (%)	7.75	7.97	5.66	-

T_1= *Trichoderma* + Rice bran + Peat soil + Water

T_2= *Trichoderma* + Wheat bran + Peat soil + Water

T_3 = *Trichoderma* + Lentil bran + Peat soil + Water

T_4 = *Trichoderma* + Gram bran + Peat soil + Water

T_5 = *Trichoderma* + Black gram bran + Peat soil + Water

T_6 = *Trichoderma* + Mustard oil cake + Peat soil + Water

T_7 = *Trichoderma* + Grass pea bran + Peat soil + Water

T_8 = *Trichoderma* + Saw dust + Peat soil + Water

T_9= Control

a

b

Plate 14. Eggplant seedlings affected by post-emergence damping off (a) 10 days after sowing (b) 13 days after sowing

4.9 Effect of different treatments on the pre-emergence damping off of eggplant seedlings

The effect of the treatments on pre-emergence damping off percentage of eggplant seedlings was presented in Table 5. All the treatments differed significantly in terms of pre-emergence damping off percentage. The highest percent pre-emergence damping off was observed in control (33.67%) followed by treatment T_8 (28.00%). The effect of treatment T_6 (*Trichoderma* + Mustered oil cake + Peat soil + Water) on pre-emergence damping off was statistically similar with the treatment T_3. The lowest percent of pre-emergence damping off (2.33%) was observed in case of treatment T_5 (*Trichoderma* + Black gram + Peat soil + Water). The second lowest percent of pre-emergence damping off (7.33%) was recorded in treatment T_7 (*Trichoderma* + Grass pea bran + Peat soil + Water) which was statistically similar with treatment T_4.

4.10 Effect of different treatments on the tip over of eggplant seedlings

The effect of the treatments on tip over of eggplant seedlings was presented in Table 5. All the treatments differed significantly in comparison to control. The highest percent tip over was observed in control (19.00%) followed by treatment T_8 (13.00%). The effect of treatment on tip over percentage (9.00%) was observed in T_6 followed by T_3 (7.00%). The lowest percent of tip over (1.33%) was recorded in treatment T_5 (*Trichoderma* + Black gram + Peat soil + Water). The second lowest percent of tip over (3.00%) was observed in case of treatment T_7 (*Trichoderma* + Grass pea bran + Peat soil + Water) preceded by treatment T_4 (*Trichoderma* + Gram bran + Peat soil + Water) (3.67%).

Table 5. Effect of different treatments on pre-emergence damping off and tip over of eggplant seedlings

Treatment	% Pre-emergence damping off	% Tip over
T_1	17.00 de	5.67 e
T_2	15.00 e	4.67 f
T_3	20.00 cd	7.00 d
T_4	10.00 f	3.67 g
T_5	2.33 g	1.33 i
T_6	22.67 c	9.00 c
T_7	7.33 f	3.00 h
T_8	28.00 b	13.00 b
T_9	33.67 a	19.00 a
$LSD_{(0.05)}$	4.29	0.66
CV (%)	14.43	5.22

T_1= *Trichoderma* + Rice bran + Peat soil + Water

T_2= *Trichoderma* + Wheat bran + Peat soil + Water

T_3 = *Trichoderma* + Lentil bran + Peat soil + Water

T_4 = *Trichoderma* + Gram bran + Peat soil + Water

T_5 = *Trichoderma* + Black gram bran + Peat soil + Water

T_6 = *Trichoderma* + Mustard oil cake + Peat soil + Water

T_7 = *Trichoderma* + Grass pea bran + Peat soil + Water

T_8 = *Trichoderma* + Saw dust + Peat soil + Water

T_9= Control

a

b

Plate 15. Eggplant seedlings affected by (a) pre-emergence damping off (b) tip over

4.11 Effect of different treatments on plant height, of eggplant seedlings at 20 days after sowing

The treatments effect on plant height of eggplant seedlings was presented in Table 6. All the treatments had significant impact on plant height of eggplant seedlings. The highest plant height was observed in case of T_5 (*Trichoderma* + Black gram + Peat soil + Water) (5.31) followed by T_7 (*Trichoderma* + Grass Pea bran + Peat soil + Water) (3.11). The third highest plant height was observed in treatment T_4 (3.77) which was statistically similar with both T_1 and T_2. The lowest plant height was observed in case of control T_9 (2.22).

4.12 Effect of different treatments on seedlings vigor of eggplant at 20 days after sowing

The treatments effect on vigor index, of eggplant seedlings was presented in Table 6. All the treatments differed significantly in terms of vigor index of eggplant seedlings. The highest vigor index (4.14) was observed in case of T_5 (*Trichoderma* + Black gram + Peat soil + Water) followed by T_7 (3.11) (*Trichoderma* + Grass Pea bran + Peat soil + Water). The vigor index (2.64) observed in case of T_4 (*Trichoderma* + Gram bran + Peat soil + Water). The effect of different treatments in vigor index was statistically similar in both T_2 and T_1. The lowest vigor index was observed in case of control T_9 (1.03).

4.13 Effect of different treatments on fresh weight of brinjal seedlings at 20 days after sowing

The treatments effect on fresh weight of brinjal seedlings was recorded and presented in Table 10. All the treatments differed significantly in terms of fresh weight of brinjal seedlings. The highest fresh weight was observed in case of T_5 (*Trichoderma* + Black gram + Peat soil + Water) (3.90gm) followed by T_7 (2.69gm). The third highest fresh weight was observed in case of T_4 (2.14gm) followed by T_2 (*Trichoderma* + Wheat bran + Peat soil + Water) (1.95gm). The lowest fresh weight was observed in case of control T_9 (1.00 gm) preceded by T_8 (*Trichoderma* + Saw dust + Peat soil + Water) (1.30gm) which was

statistically identical with T_6 (*Trichoderma* + Mustared oil cake + Peat soil + Water) (1.37gm).

Table 6. Effect of different treatments on plant height, vigor index and fresh weight of eggplant seedlings at 20 days after sowing

Treatment	Plant height (cm)	Vigor index	Fresh weight (gm)
T_1	3.50 cd	2.21 de	1.78 e
T_2	3.60 cd	2.34 d	1.95 d
T_3	3.33 de	2.00 e	1.53 f
T_4	3.77 c	2.64 c	2.14 c
T_5	5.31 a	4.14 a	3.90 a
T_6	3.04 e	1.74 f	1.37 g
T_7	4.28 b	3.11 b	2.69 b
T_8	2.71 f	1.41 g	1.30 g
T_9	2.22 g	1.03 h	1.00 h
$LSD_{(0.05)}$	0.31	0.21	4.55
CV (%)	5.04	5.36	0.15

T_1= *Trichoderma* + Rice bran + Peat soil + Water

T_2= *Trichoderma* + Wheat bran + Peat soil + Water

T_3 = *Trichoderma* + Lentil bran + Peat soil + Water

T_4 = *Trichoderma* + Gram bran + Peat soil + Water

T_5 = *Trichoderma* + Black gram bran + Peat soil + Water

T_6 = *Trichoderma* + Mustard oil cake + Peat soil + Water

T_7 = *Trichoderma* + Grass pea bran + Peat soil + Water

T_8 = *Trichoderma* + Saw dust + Peat soil + Water

T_9= Control

Plate 16. Seedling showing the highest plant height, vigor index and fresh weight of eggplant raised on treatment T_5 (*Trichoderma* + Black gram bran + Peat soil + Water)

Plate 17. Seedling showing the lowest plant height, vigor index and fresh weight of eggplant raised on control pot

4.14 Effect of different treatments on the germination of tomato seedlings at 7 days after sowing

The treatments effect on germination of tomato seedlings at different days after sowing (DAS) was presented in Table 7. The effect of different treatments on germination of tomato seedlings at 7 DAS differed significantly in comparison to control. The highest germination (81.11%) was observed in T_5 (*Trichoderma* + Black gram + Peat soil + Water) and the lowest germination (48.89%) was observed in T_9 (Control). The second highest germination (76.67%) was observed in treatment T_7 (*Trichoderma* + Grass pea bran + Peat soil + Water) which was statistically similar with T_4 (74.08%). The effect of treatment T_2 on germination at 7 DAS was statistically similar with the effect of treatment T_1 which was also statistically similar with treatment T_3 (*Trichoderma* + Lentli bran + Peat soil + Water).

4.15 Effect of different treatments on the germination of tomato seedlings at 10 days after sowing

A remarkable effect was observed among the treatments on the germination at 10 DAS of tomato seedling. The effect of different treatments on germination of tomato seedlings differed significantly in comparison to control. At 10 DAS, the highest germination (82.41%) was observed in T_5 (*Trichoderma* + Black gram + Peat soil + Water) and the lowest germination (50.19%) was observed in T_9 (Control). The second highest germination (77.96%) was observed in T_7 (*Trichoderma* + Grass pea bran + Peat soil + Water) which was statistically similar with treatment T_4 (75.18%). The effect of treatment T_2 on germination at 10 DAS was statistically similar with the effect of treatment T_4 and T_1. The effect of treatment T_1 on germination at 10 days after sowing was also statistically similar with treatment T_6.

4.16 Effect of different treatments on the germination of tomato seedlings at 13 days after sowing

The effect of different treatments on germination of tomato seedlings differed significantly in comparison to control. At 13 DAS, the highest germination (83.15%) was observed in T_5 (*Trichoderma* + Black gram + Peat soil + Water) and the lowest germination (51.11%) was observed in T_9 (Control). The second highest germination (79.07%) at the same days after sowing was observed in T_7 (*Trichoderma* + Grass pea bran + Peat soil + Water) which was statistically similar with treatment T_4 (76.30%). The effect of treatment T_1 on germination at 13 DAS was statistically similar with the effect of treatment T_2 and treatment T_6 (*Trichoderma* + Mustered oil cake + Peat soil + Water). According to the performance of the treatments the highest increase of seed germination (62.69%) was counted in T_5 (*Trichoderma* + Black gram + Peat soil + Water) followed by T_7 (54.70%) (*Trichoderma* + Grass pea bran + Peat soil + Water), T_4 (49.28%) (*Trichoderma* + Gram bran + Peat soil + Water) and T_2 (44.19%) (*Trichoderma* **+** Wheat bran + Peat soil + Water) respectively.

Table 7. Effect of different treatments on the germination of tomato seedlings at different days after sowing (DAS)

Treatment	% Germination			% germination increased over control
	7 DAS	10 DAS	13 DAS	13 DAS
T_1	69.07 de	70.00 de	71.67 de	40.23
T_2	71.48 cd	72.59 cd	73.70 cd	44.19
T_3	67.04 ef	69.07 de	70.00 e	36.96
T_4	74.08 bc	75.18 bc	76.30 bc	49.28
T_5	81.11 a	82.41 a	83.15 a	62.69
T_6	65.00 f	66.85 e	69.07 e	35.14
T_7	76.67 b	77.96 b	79.07 b	54.70
T_8	54.07 g	55.92 f	56.66 f	10.85
T_9	48.89 h	50.19 g	51.11 g	-
$LSD_{(0.05)}$	3.11	3.49	3.09	-
CV (%)	2.68	2.95	2.57	-

T_1= *Trichoderma* + Rice bran + Peat soil + Water

T_2= *Trichoderma* + Wheat bran + Peat soil + Water

T_3 = *Trichoderma* + Lentil bran + Peat soil + Water

T_4 = *Trichoderma* + Gram bran + Peat soil + Water

T_5 = *Trichoderma* + Black gram bran + Peat soil + Water

T_6 = *Trichoderma* + Mustard oil cake + Peat soil + Water

T_7 = *Trichoderma* + Grass pea bran + Peat soil + Water

T_8 = *Trichoderma* + Saw dust + Peat soil + Water

T_9= Control

a

b

Plate 18. Showing healthy seedlings of tomato raised on treatment T_5 (*Trichoderma* + Black gram bran + Peat soil + Water) (a) 10 days after sowing (b) 13 days after sowing

4.17 Effect of different treatments on the post-emergence damping off of tomato seedlings at 7 days after sowing

Treatments effect differed significantly in respect of post-emergence damping off percentage at 7 DAS (Table 8). The highest percent post-emergence damping off was recorded in control (14.44%). The second highest post-emergence damping off (12.22%) at 7 DAS was observed in treatment T_8 (*Trichoderma* + Saw dust + Peat soil + Water). The effect of treatment T_3 on post-emergence damping off at 7 DAS was statistically similar with the treatment T_6 (*Trichoderma* + Mustered oil cake + Peat soil + Water). The lowest percent of post-emergence damping off (1.11%) was recorded in case of treatment T_5 (*Trichoderma* + Black gram + Peat soil + Water). The second lowest percent of post-emergence damping off (3.33%) was observed in case of treatment T_7 (*Trichoderma* + Grass Pea bran + Peat soil + Water) preceded by T_4 (5.18%).

4.18 Effect of different treatments on the post-emergence damping off of tomato seedlings at 10 days after sowing

A remarkable effect was observed among the treatments in controlling post-emergence damping off disease of tomato at 10 DAS (Table 8). The treatments effects differed significantly in comparison to control. The highest percent post-emergence damping off was recorded in control (15.89%). The second highest post-emergence damping off (13.00%) at 10 DAS was observed in treatment T_8. The effect of treatment T_3 on post-emergence damping off at 10 DAS was statistically similar with the treatment T_6. The lowest post-emergence damping off (2.22%) was recorded in case of treatment T_5 (*Trichoderma* + Black gram + Peat soil + Water). The second lowest post-emergence damping off (4.26%) was observed in case of treatment T_7 (*Trichoderma* + Grass pea bran + Peat soil + Water) preceded by T_4 (6.67%).

4.19 Effect of different treatments on the post-emergence damping off of tomato seedlings at 13 days after sowing

The effect of the treatments on post-emergence damping off percentage of tomato seedlings at 13 days after sowing (DAS) was presented in Table 8. All the treatments differed significantly in terms of post-emergence damping off percentage. The highest percent post-emergence damping off was recorded in control (17.55%). The second highest post-emergence damping off (14.44%) at same DAS was observed in treatment T_8. The effect of treatment T_3 on post-emergence damping off at 13 DAS was statistically similar with the treatment T_6. The lowest percent of post-emergence damping off (3.33%) was recorded in case of treatment T_5 (*Trichoderma* + Black gram + Peat soil + Water). The second lowest percent of post-emergence damping off (5.55%) was observed in case of treatment T_7. The third lowest percent of post-emergence damping off (7.41%) was observed in case of treatment T_4 (*Trichoderma* + Gram bran + Peat soil + Water). According to the performance of the treatments the highest reduction of post-emergence damping off (81.02%) was counted in T_5 (*Trichoderma* + Black gram + Peat soil + Water) followed by T_7 (68.37%) (*Trichoderma* + Grass pea bran + Peat soil + Water), T_4 (57.78%) (*Trichoderma* + Gram bran + Peat soil + Water) and T_2 (49.34%) (*Trichoderma* + Wheat bran + Peat soil + Water) respectively.

Table 8. Effect of different treatments on post-emergence damping off of tomato seedlings at different days after sowing (DAS)

Treatment	% Post-emergence damping off			% post-emergence damping off reduced over control
	7 DAS	10 DAS	13 DAS	13 DAS
T_1	7.96 d	9.81 cd	9.99 cd	43.07
T_2	6.85 e	8.70 d	8.89 d	49.34
T_3	9.07 c	10.92 c	11.11 c	36.69
T_4	5.18 f	6.67 e	7.41 e	57.78
T_5	1.11 h	2.22 g	3.33 g	81.02
T_6	9.07 c	10.74 c	11.11 c	36.69
T_7	3.33 g	4.26 f	5.55 f	68.37
T_8	12.22 b	13.00 b	14.44 b	17.72
T_9	14.44 a	15.89 a	17.55 a	-
$LSD_{(0.05)}$	0.88	1.52	1.41	-
CV (%)	6.64	9.73	8.30	-

T_1= *Trichoderma* + Rice bran + Peat soil + Water

T_2= *Trichoderma* + Wheat bran + Peat soil + Water

T_3 = *Trichoderma* + Lentil bran + Peat soil + Water

T_4 = *Trichoderma* + Gram bran + Peat soil + Water

T_5 = *Trichoderma* + Black gram bran + Peat soil + Water

T_6 = *Trichoderma* + Mustard oil cake + Peat soil + Water

T_7 = *Trichoderma* + Grass pea bran + Peat soil + Water

T_8 = *Trichoderma* + Saw dust + Peat soil + Water

T_9= Control

a

b

Plate 19. Showing tomato seedlings affected by post-emergence damping off (a) 7 days after sowing (b) 10 days after sowing

4.20 Effect of different treatments on the pre-emergence damping off of tomato seedlings

The effect of the treatments on pre-emergence damping off percentage of tomato seedlings was presented in Table 9. All the treatments differed significantly in respect of pre-emergence damping off percentage. The highest percent pre-emergence damping off was observed in control (33.89%) followed by treatment T_8 (*Trichoderma* + Saw dust + Peat soil + Water) (28.33%). The effect of treatment T_6 on pre-emergence damping off was (20.19%). The effect of treatment T_1 (*Trichoderma* + Rice bran + Peat soil + Water) on pre-emergence damping off was statistically similar with treatment T_3 (*Trichoderma* + Lentli bran + Peat soil + Water) and T_2 (*Trichoderma* + Wheat bran + Peat soil + Water). The lowest percent of pre-emergence damping off (1.85%) was observed in case of treatment T_5 (*Trichoderma* + Black gram + Peat soil + Water). The second lowest percent of pre-emergence damping off (5.92%) was recorded in treatment T_7 (*Trichoderma* + Grass pea bran + Peat soil + Water).

4.21 Effect of different treatments on the tip over of tomato seedlings

The effect of the treatments on tip over percentage of eggplant seedlings was presented in Table 9. All the treatments differed significantly in terms of tip over percentage. The highest percent tip over was observed in control (10.00%) followed by treatment T_8 (*Trichoderma* + Saw dust + Peat soil + Water) (6.67%). The effect of treatment on tip over percentage (4.44%) was observed in T_6 followed by T_3 (3.33%). The lowest percent of tip over (0.55%) was recorded in treatment T_5 (*Trichoderma* + Black gram + Peat soil + Water). The second lowest percent of tip over (1.11%) was observed in case of treatment T_7 (*Trichoderma* + Grass pea bran + Peat soil + Water) followed by treatment T_4 (1.85%).

Table 9. Effect of different treatments on pre-emergence damping off and tip over of tomato seedlings

Treatment	% Pre-emergence damping off	% Tip over
T_1	15.18 d	2.78 e
T_2	12.41de	2.41 f
T_3	15.00 d	3.33 d
T_4	11.30 e	1.85 g
T_5	1.85 g	0.55 i
T_6	20.19 c	4.44 c
T_7	5.92 f	1.11 h
T_8	28.33 b	6.67 b
T_9	33.89 a	10.00 a
$LSD_{(0.05)}$	3.27	0.26
CV (%)	11.90	4.10

T_1= *Trichoderma* + Rice bran + Peat soil + Water

T_2= *Trichoderma* + Wheat bran + Peat soil + Water

T_3 = *Trichoderma* + Lentil bran + Peat soil + Water

T_4 = *Trichoderma* + Gram bran + Peat soil + Water

T_5 = *Trichoderma* + Black gram bran + Peat soil + Water

T_6 = *Trichoderma* + Mustard oil cake + Peat soil + Water

T_7 = *Trichoderma* + Grass pea bran + Peat soil + Water

T_8 = *Trichoderma* + Saw dust + Peat soil + Water

T_9= Control

a

b

Plate 20. Tomato seedlings affected by (a) pre-emergence damping off and (b) tip over

4.22 Effect of different treatments on plant height of tomato seedlings at 16 days after sowing

All the treatments differed significantly in terms of plant height of tomato seedlings. The highest plant height (11.4 cm) was observed in case of T_5 (*Trichoderma* + Black gram + Peat soil + Water) followed by T_7 (8.33 cm) (*Trichoderma* + Grass pea bran + Peat soil + Water). The third highest plant height (7.56 cm) was observed in case of T_4 (*Trichoderma* + Gram bran + Peat soil + Water) which was statistically identical with the treatment T_2 (6.87 cm). The lowest plant height was observed in case of control T_9 (3.29 cm). The second lowest plant height (4.15 cm) was observed in T_8 (*Trichoderma* + Saw dust + Peat soil + Water) which was statistically similar with T_6 (4.78 cm). The effect of different treatments in plant height was statistically similar in both T_3 and T_1.

4.23 Effect of different treatments on vigor index of tomato seedlings at 16 days after sowing

The treatments effect on vigor index, of tomato seedlings was presented in Table 10. All the treatments differed significantly in terms of vigor index of tomato seedlings. The highest vigor index was observed in case of T_5 (*Trichoderma* + Black gram + Peat soil + Water) (9.49) followed by T_7 (*Trichoderma* + Grass Pea bran + Peat soil + Water) (6.59). The third highest vigor index was observed in case of T_4 (5.76) followed by treatment T_2 (5.06). The effect of different treatments in vigor index was statistically similar in both T_1 and T_3. The lowest vigor index was observed in case of control T_9 (1.68). The second lowest vigor index was observed in T_8 (2.35) preceded by T_6 (3.30).

4.24 Effect of different treatments on fresh weight of tomato seedlings at 16 days after sowing

The treatments effect on fresh weight of tomato seedlings was presented in Table 10. All the treatments effect differed significantly in terms of fresh

weight of tomato seedlings. The highest fresh weight (8.20 gm) was observed in case of T_5 (*Trichoderma* + Black gram + Peat soil + Water) followed by T_7 (6.23 gm) (*Trichoderma* + Grass Pea bran + Peat soil + Water). The third highest fresh weight (5.17 gm) was observed in case of T_4 (*Trichoderma* + Gram bran + Peat soil + Water) which was statistically similar with T_2 (4.90 gm). The lowest fresh weight was observed in case of control T_9 (2.23 gm). The second lowest fresh weight was observed in T_8 (3.23 gm) preceded by T_6 (3.83 gm).

Table 10. Effect of different treatments on plant height, vigor index and fresh weight of tomato seedlings at 16 days after sowing

Treatment	Plant height (cm)	Vigor index	Fresh weight (gm)
T_1	5.93 d	4.25 e	4.70d
T_2	6.87 c	5.06 d	4.90cd
T_3	5.99 d	4.19 e	4.17e
T_4	7.56 c	5.76 c	5.17c
T_5	11.41a	9.49 a	8.20a
T_6	4.78 e	3.30 f	3.83f
T_7	8.33 b	6.59 b	6.23b
T_8	4.15e	2.35 g	3.23g
T_9	3.29 f	1.68 h	2.23h
$LSD_{(0.05)}$	0.69	0.54	0.30
CV (%)	6.21	6.61	3.72

T_1= *Trichoderma* + Rice bran + Peat soil + Water

T_2= *Trichoderma* + Wheat bran + Peat soil + Water

T_3 = *Trichoderma* + Lentil bran + Peat soil + Water

T_4 = *Trichoderma* + Gram bran + Peat soil + Water

T_5 = *Trichoderma* + Black gram bran + Peat soil + Water

T_6 = *Trichoderma* + Mustard oil cake + Peat soil + Water

T_7 = *Trichoderma* + Grass pea bran + Peat soil + Water

T_8 = *Trichoderma* + Saw dust + Peat soil + Water

T_9= Control

Plate 21. Showing the highest plant height, vigor index and fresh weight of tomato seedlings raised on treatment T_5 (*Trichoderma* + Black gram bran + Peat soil + Water)

Plate 22. Showing the lowest plant height, vigor index and fresh weight of tomato seedlings raised on control pot

DISCUSSION

The present study was carried out with nine different treatments of *Trichoderma* based substrates viz. T_1 (*Trichoderma* + Rice bran + Peat soil + Water), T_2 (*Trichoderma* + Wheat bran + Peat soil + Water), T_3 (*Trichoderma* + Lentil bran + Peat soil + Water), T_4 (*Trichoderma* + Gram bran + Peat soil + Water), T_5 (*Trichoderma* + Black gram bran + Peat soil + Water), T_6 (*Trichoderma* + Mustard oil cake + Peat soil + Water), T_7 (*Trichoderma* + Grass Pea bran + Peat soil + Water), T_8 (*Trichoderma* + Saw Dust + Peat soil + Water), T_9 (Control) were evaluated for sporulations of *Trichoderma harzianum* and acting against *Sclerotium rolfsii* for the management of damping off of eggplant and tomato seedlings.

Different isolates of *Trichoderma* evaluated in the experiment found to be differed in terms of number of spore production. The highest number of spore ($6.42 \times 10^4/mm^2$) was observed in isolate T_1 (TJG - *Trichoderma* sp. Joydevpur, Gazipur) that was used in the substrate to construct the treatment combinations.

In mass multiplication of *Trichoderma* isolate TJG (*Trichoderma* sp. Jodevpur, Gazipur) black gram brans combined with peat soil and water (1:1:2 w/w/v) proved to be suitable substrate for producing the highest CFU/g of *Trichoderma* sp. Earlier studies support the results of the present findings (Shamsuzzaman *et.al*., 2003a, Islam, 2005). Shamsuzzaman *et.al*., 2003 reported that the highest production conidia (42.93×10^7/g) was recorded in black gram based substrates while used for mass multiplication of *Trichoderma* isolates. Several other workers also used different agro-products for multiplication of *Trichoderma sp*. but they did not include peat soil based black gram brans in their study (Rettinassababady *et.al.,* 2000, Shamarao *et. al.,* 1998).

The peat soil based Trichodermal formulations were evaluated in the nursery house against *Sclerotium rolfsii* for the management of eggplant and tomato seedlings. The data recorded on percent seed germination, post-emergence damping off, pre-emergence damping off, tip over, plant height and vigor index of at different days after sowing (DAS). In case of eggplant seedlings, the results revealed that the treatment T_5 (*Trichoderma* + Black gram bran + Peat soil + Water) and T_7 (*Trichoderma* + Grass Pea bran + Peat soil + Water) had remarkable effect against damping off and tip over of seedlings increasing the seed germination, seedling height, vigor index and fresh weight of seedlings irrespective of days after sowing (DAS). At 16 DAS, the highest germination (78.00%) was observed in T_5 (*Trichoderma* + Black gram bran + Peat soil + Water) followed by T_7 (*Trichoderma* + Grass Pea bran + Peat soil + Water).

In case of post-emergence damping off at 16 DAS, the highest reduction (78.90%) of post-emergence damping off was observed in case of treatment T_5 (*Trichoderma* + Black gram bran + Peat soil + Water) followed by T_7 (*Trichoderma* + Grass Pea bran + Peat soil + Water). Same was the case in pre-emergence damping off seedlings where reduction of pre-emergence damping was observed in case of treatment T_5 (*Trichoderma* + Black gram bran + Peat soil + Water) followed by T_7 (*Trichoderma* + Grass Pea bran + Peat soil + Water). In case of plant height, the highest plant height (5.31 cm) was observed in T_5 (*Trichoderma* + Black gram bran + Peat soil + Water) and the second highest highest plant height (4.28 cm) was observed in T_7 (*Trichoderma* + Grass Pea bran + Peat soil + Water).

In case of vigor index the highest vigor index (4.14) was observed in T_5 (*Trichoderma* + Black gram bran + Peat soil + Water) and the second highest highest vigor index (3.11) was observed in T_7 (*Trichoderma* + Grass Pea bran + Peat soil + Water). In case of fresh weight of seedlings, the highest fresh weight (3.90 gm) was observed in T_5 (*Trichoderma* + Black gram bran + Peat soil + Water) while the second highest fresh weight (2.69 gm) was observed in T_7 (*Trichoderma* + Grass Pea bran + Peat soil + Water). The performance of

the peat soil based Trichodermal substrates in case of the tomato seedlings, were more or less similar to that of eggplant seedlings for the management of damping off of seedlings,

The present findings were keep in with the findings of Meah *et al*. (2004) who reported that *Trichoderma harzianum* cp and *Trichoderma harzianum* T_{22} grown on peat soil based black gram bran was found effective in controlling nursery diseases like damping off , tip over and seedling blight of eggplant and promoted seed germination. Shamsuzzaman *et al*. (2003a) studied for mass production of *Trichoderma harzianum*. Of them, rice straw chick pea bran, rice course with 3% chickpea powder, rice straw with 5% sucrose black gram bran, grass pea bran and peat based wheat bran supported best in mass production of conidia (42.93×10^7/g culture). Shamsuzzaman *et al*. (2003b) further reported that seed treatment with *Trichoderma harzianum* grown on black gram resulted up to 16.66% higher seed germination , 266.33% fresh shoot weight, 157.14% fresh root weight and 98.55 vigor index of cucurbits over control. Shahiduzzaman (2009) found that *T. harzianum* present in decomposed municipality waste was effective in increased growth response by increasing number of leaves. Ozbay *et al*. (2004) and Mukhopodhyay (1989) demonstrated increased growth of several crop plants in the presence of biological agents. These responses may be caused by a direct effect to the plant (as bio-fertilizer) or by control of some undiagnosed plant pathogens. Increase plant height, fresh and dry weight of wheat (1.5 times), cucumber (75.2 times), and radish (1.9 times) were achieved when plant growth promoting fungi (PGPF) *Trichoderma* were applied in the soil. Abd-El-Khair *et al.* (2010) found that the average of bean plant height with *Trichoderma* application were in the range of 46.0-49.8 cm compared to 37.3 cm in the control plants and also found that the average fresh weight of pods in bean were in the range of 43.1-77.4% g in case of *Trichoderma* treated, compared to 42.5g in the control plants. Currently, the role of biological control agents is a well established fact and has become increasingly crucial, and in several cases, complementary or even replacing the chemical counterparts where antagonistic fungi play an important

role (Whipps and Lumsden, 2001; Chet, 1993). In this context, *Trichoderma* spp. have been the cynosure of many researchers who have been contributing to biological control pursuit through use of fungi (Heraux *et al*., 2005a; Heraux *et al*., 2005b; Ortiz and Orduz, 2001). Furthermore, *Trichoderma* spp. share almost 50% of fungal Biological Control Agents market, mostly as soil/growth enhancers and this makes them interesting candidates to investigate (Whipps and Lumsden, 2001).

SUMMARY AND CONCLUSION

The experiments were conducted in the laboratory as well as in the nursery house of the Department of Plant Pathology, Sher-e-Bangla Agricultural University, Dhaka, during the period from December 2013 to September 2014. The experiment was carried out with nine different treatments viz. T_1 (*Trichoderma* + Rice bran + Peat soil + Water), T_2 (*Trichoderma* + Wheat bran + Peat soil + Water), T_3 (*Trichoderma* + Lentil bran + Peat soil + Water), T_4 (*Trichoderma* + Gram bran + Peat soil + Water), T_5 (*Trichoderma* + Black gram bran + Peat soil + Water), T_6 (*Trichoderma* + Mustard oil cake + Peat soil + Water), T_7 (*Trichoderma* + Grass Pea bran + Peat soil + Water), T_8 (*Trichoderma* + Saw Dust + Peat soil + Water) and T_9 (Control). Data were recorded on the germination, post-emergence damping off, pre-emergence damping off, tip over, plant height, vigor index and fresh weight of eggplant and tomato seedlings. The data were analyzed by using MSTAT statistical package program.

Trichoderma harzianum was isolated from different agro-ecological areas of Bangladesh. Among them (TJG – *Trichoderma harzianum*, Joydevpur, Gazipur) was the best isolate in respect of number spore production (6.42×10^4). Different substrates were used with peat soil and water at a ratio (1:1:2) to know the inoculum potentiality in respect of number spore of *Trichoderma harzianum*. The highest number of spore/gm (24.27×10^7) was observed in (Black gram bran + peat soil + water, at ratio - 1:1:2).

The *Trichoderma* based biopesticide had significant effects on all the parameters studied. All the parameter showed good result when T_5 (*Trichoderma* + Black gram bran + Peat soil + Water) was applied. The control treatment gave poor result in respect of all the parameter.

Irrespective of data taken in different days after sowing. The highest germination of eggplant seedlings was observed in T_5 (*Trichoderma* + Black gram bran + Peat soil + Water) and second highest germination was observed

in T_7 (*Trichoderma* + Grass Pea bran + Peat soil + Water). At 10 DAS, the highest germination (76.00%) was observed in T_5. At 16 DAS, the highest germination (78.00%) was observed in T_5 and second highest germination (72.67%) was observed in T_7. At 10 DAS, 13 DAS and 16 DAS the lowest post-emergence damping off of eggplant seedlings was observed in T_5 (*Trichoderma* + Black gram bran + Peat soil + Water) and second lowest post-emergence damping off was observed in T_7. The lowest pre-emergence damping off and tip over of eggplant seedlings were also observed in T_5 and the second lowest pre-emergence damping off and tip over were observed in T_7. The highest plant height, vigor index and fresh weight of eggplant seedlings were observed in T_5 followed by T_7.

In case of tomato seedlings, the highest germination (81.11%) at 7 DAS was observed in T_5 and the second highest germination (76.67%) was observed in T_7. At 10 DAS, the highest germination (82.41%) was observed in T_5 and second highest germination (77.96%) was observed in T_7. At 13 DAS the highest germination of tomato seedling was 83.15% counted in treatment T_5 followed by treatment T_7 (79.07%). At 7 DAS, 10 DAS and 13 DAS the lowest post-emergence damping off of tomato seedlings was observed in T_5 (1.11%, 2.22% and 3.33% respectively) and second lowest post-emergence damping off was observed in T_7 (3.33%, 4.26% and 5.55% respectively). The lowest pre-emergence damping off (1.85%) and tip over (0.55%) of tomato seedlings were observed in T_5 and second lowest pre-emergence damping off (5.92%) and tip over (1.11%) were observed in T_7. The highest plant height, vigor index and fresh weight of tomato seedlings were also observed in T_5 followed by T_7.

REFERENCES

Abd-El-Khair, H., Khalifa, R.Kh.M. and Karima, H.E. Haggag. (2010). Effect of *Trichoderma* species on damping off diseases incidence, some plant enzymes activity and nutritional status of bean plants. *J. Amer.Sci.* **6**(9):486-497.

Agrawal, S.C., Khare, M.N. and Agrawal, P.S. (1977). Biological control of *Sclerotium rolfsii* causing collar rot of lentil. **30**(2): 176-179.

Ahmed, H.U. and Hossain, M.M. (1985). Establishment of a disesese harberium in Bangladesh Agricultural Research Institute (BARI). Final Report, Plant Pathology Division, BARI Joydevpur, Gazipur 1701, Bangladesh.

Akhtar, C. M. (1977). Biological control of some plant diseases lacking genetic resistance of the host crops in Pakistan. *Ann. N. Y. Acad. Sci.* 187: 45.

Amin, F., Razdanl, V.K., Mohiddin, F.A., Bhat, K.A. and Banday, S. (2010). Potential of *Trichoderma* species as Bio control Agents of Soil Borne Fungal Propagules. *J. Phytol.* **2**(10): 38–41.

Anonymous. (1992). BARI (Bangladesh Agricultural Research Institute) Annual report (1991-1992), BARI, Joydebpur, Gazipur. 74p.

Ashrafuzzaman, H.A. (1976). Labortory manual of plant pathology. First edition. Department of Plant Pathology, Bangladesh Agricultural University, Mymensingh. 94 p.

Babar, H.M. 1999. Studies on collar rot of sunflower. Ph.D. thesis, Dept. Plant Pathology, Bangladesh Agricultural University, Mymensingh, Bangladesh. 153 p.

Backman, P.A. and Rodriguez-Kabana, R.A. (1975). A system for the growth and delivery of biological control agents to the soil. *Phytopathology*. **65**: 819.

Baker, R. (1988). *Trichoderma* spp. As plant growth stimulants. CRC Critical Rev. Biotech. **7**: 97-106.

BBS. (2008). Year Book of Agricultural Statistics of Bangladesh. Bangladesh Bureau of Statistics, Planning Division, Ministry of Planning, Govt. of the Peoples Republic of Bangladesh, Dhaka. p.106.

BBS. (2011). Year Book of Agricultural Statistics of Bangladesh. Bangladesh Bureau of Statistics, Planning Division, Ministry of Planning, Govt. of the Peoples Republic of Bangladesh, Dhaka. p.108.

Begum, M.M. (1997). Efficacy of *Trichoderma harzianum* as a biocontrol agent for controlling *Fusarium* spp. and *Sclerotium rolfsii* in food legumes. MS thesis, Dept. of Plant Pathology. Bangladesh Agricultural University, Mymensingh. Bangladesh. P.61.

Bose, T.K. and Som, M.G. (1986). Vegetables of India, Nayaprokash Calcutta, India pp.312-313.

Burr, T.J., Schroth, M.M. and Suslow, T. (1978). Increased potato yields by treatment of seed pieces with specific strains of *Pseudomonas flurescens* and *P. putida*. *Phytopathlogy.* **68**: 1378-1385.

Chandrasehar, G., Ayyappan, S. and Eswaran, A. (2005). Management of tomato collar rot caused by *Sclerotium rolfsii* by antagonistic microorganisms. *J. Ecobiology*. 17(3): 261-264.

Chet, I. (1993). Biotechnology in Plant Disease Control, John Wiley and Sons, New York.

Chet, I., and Inbar, A. (1994). Genetic diversity and vegetative compatibility among *T. harzianum* isolates. *Molecular General Genetics.* **256**: 127-135.

Chowdhury, M.S.M., Hossain, I., Fakir, G.A., Aminuzzaman, F.M. and Islam, M.R. (2000). Tolerance of *Trichoderma harzianum* and *Gliocladium viride* to agrochemicals and their antagonistic effect on seed born mycoflora of Pigeon Pea. *Bangladesh J. Seed Sci. & Tech*. **4**(1&2): 83-86.

Churchill, W.B. (1982). Mass production of microorganisms for biological control. In Biological control of weeds with Plant Pathogens. Charudattan, R. and walker, H. L. Eds., Jhon Wiley and Sons, New York. 139.

Das, A.C. (1984). Effect of cultural practices on the damping off incidence of TPS seedlings and it's chemical control in vitro. M.Sc.Ag. Thesis, Bangladesh Agricultural University. 50p.

Das, B.C., Dutta, P., Devi, G. and Dutta, P. (2000). Management of *Sclerotium rolfsii* in tomato by fungal antagonists. *J. Agril. Sci.* Society of North East India. **13**(1): 101-103.

Das, B.C., Roy, S.K. and Bora, L.C. (1997). Mass multiplication of *Trichoderma* species on different media. *J. Agril. Sci*. Society of North East India. **10**(1): 95-100.

Dasgupta, M.K. (1988). Principles of Plant Pathology. Allied Publisher Privet Limited. New Delhi, India. 700p.

Dhingra, O.D. and Sinclair, J.B. (1985). Basic Plant Pathology Methods. CRC Press, Boca Raton, Inc, FL. 355Pp.

Dubey, S.C. and Patel, B. (2002). Mass multiplication of antagonist and standardization of effective dose for management of web blight of urd and mung bean. *Indian Phytopatholol*. **55**(3):338-341.

Elad, Y., Chet, I. and Katan, J. (1982). *Trichoderma harzianum*: A biocontrol agent effective against *Sclerotium rolfsii* and *Rhizoctonia solani*. Phytopathology. **70**: 119-121.

Elango, F. (1986). A simple greenhouse incubation techniques for screening true potato seedlings for their tolerance to *Rhizoctonia solani* induced damping off. *Phytopathology*. 59: 466-467.

Fokkema, N.J. (1995). Strategies for biocontrol of foliar fungal diseases. In M. Manka (ed.), Environmental Biotic Factors in Integrated Plant Disease Control. The Polish Phytopathological Society. Poznan. pp. 69-79.

Gowda, C.L.L. and Kaul, A.K. (1982). Nutritional and antinutrional consideration. Pulses in Bangladesh. 431p.

Hader, Y., Chet, I. and Hanis. Y. (1979). Biological control of *Rhizoctonia solani* damping off with wheat bran culture of *Tricoderma harzianum. Phytopathology*. **69**: 64-68.

Haque, E.S.A., Ghaffar, A. and Zaki, M.J. (1990). Biological control of root rot disease of okra, subflower, soybean and mungbean. *Pakistan J. Botany.* **22**(2): 212-214.

Haque, M.E. (2006). Study of Genetic Diversity of different Brinjal (*Solanum melongena* L) varieties and Lines by RAPD markers. An M.S. thesis submitted to the Dept. of Biotechnology. Bangladesh Agricultural University, Mymensingh. PP. 30-42.

Heraux, F.M.G., Hallett, S.G., Ragothama, K.G. and Weller, S.C. (2005a). Composted chicken manure as a medium for the production and delivery of *Trichoderma virens* for weed control. *Hort. Science*. **40**: 1394–1397.

Heraux, F.M.G., Hallett, S.G., Ragothama, K.G. and Weller, S.C. (2005b). Combining *Trichoderma virens*-inoculated compost and a rye cover crop for weed control in transplanted vegetables. *Biol. Control*. **34**: 21–26.

Hossain, I. and Hossain, M.H. (2010). Status of tikka disease of groundnut in Bangladesh and effect of BAU-Biofungicide on improving seed quality. *BAU Res. Prog*. **20**: 29-30.

Howlader, M. (2003). Plant growth promoting fungi from Trufgrass rhizosphere with potential for disease suppression soil organisms. **44**: 33-38.

Inbar, J., Abramsky. M., Cohen, D. and Chet, I. (1994). Plant growth enhancement and disease control by *Trichoderma harzianum* in vegetable seedlings grown under commercial conditions. *European J. Pl. Pathol*. **1000**: 337-346.

Iqbal, S.M., Bakhsh, A. Hussain, S. and Malik, B.A. (1995). Microbial antagonism against *Sclerotium rolfsii*, the cause of collar rot of lentil. *Lens Newsletter*. **22** (1/2): 48-49.

Islam, M.N., Bhuyan, M.K.A., Mian, I.H. and Bari, M.A. (2002). Evaluation of nine organic substrates for mass culture of *Trichoderma harzianum*. *Bangladesh J. Plant Pathol.* **18**(1&2): 43-48.

Islam, M.R. (2005). An integrated approach for management of phomopsis blight and fruit rot of eggplant. Ph.D. thesis, Dept. of Plant Pathology. Bangladesh Agricultural University, Mymensingh, Bangladesh.

Kashem, A. (2005). *Trichoderma* in controlling foot and root rot and collar rot of lentil. Ph.D. thesis, Dept. of Plant Pathology, Bangladesh Agricultural University, Myensingh, Bangladesh. 192 pp.

Kashem, M.A., Hossain, I. and Hasna, M.K. (2011). Use of *Trichoderma* in biological control of foot and root rot of lentil (*Lens culinaris* Medik). *Int. J. Sustain. Crop Prod*. **6**(1), 29-35.

Kaur, N.P. and Mukhapadhyay, A.N. (1992). Integrated control of chickpea with complex by *Trichoderma* and chemical methods in India. *Tropical Pest management*. **38**(4): 372-375.

Kenny, D.S. and Couch, T.L. (1981). Mass production of biological control agents for plant diseases, weeds and insects control. In Biological control in crop production. Papavizas, G.C. Ed. Allenheld, *Totowa, N.J*. 143p.

Klean, H. and Murugesan, K. (2003). Hyperparasitic potential *Trichoderma harzianum* over *Fusarium* sp. vasinfectum. *Indian J. Microbiol*. **36**(3): 145-148.

Krishnamoorthy, A.S. and Bhoskaran, R. (1990). Biological control of damping off disease of tomato caused by *Pythium indicum*. *Balakrisham. J. of biological control*. **4**(1): 52-54.

Kumar, S.M. and Khare, M.N. (1990). Studies on the antagonistic relationship of soybean spermosphere microflora with *Rhizoctonia bataticola* and *Sclerotium rolfsii*. *J. Biological control*. **4**(1): 72-74.

Kumar. S. (2013). Trichoderma: a biological weapon for managing plant diseases and promoting sustainability. *Int. J. Agrl. Sc. & Vet. Med*. Vol. 1, No. 3.

Kwok, O.C.H., Fahy, P.C., Hoitink, H.A.J. and Kuter, G.A. (1987). Interaction between bacteria and *Trichoderma hamatum* in suppression of *Rhizoctonia* damping-off in bark compost media. *Phytopath.* **77**(8): 1206-1212.

Lewis, J.A. and Pavavizas, G.C. (1980). Integrated control of *Rhizoctonia* fruit rot of cucumber. *Phytopathology*. 70: 70-85.

Mangenot, F. and Diem, H.G. (1979). Fundamental of biological control, in ecology of root pathogen Krupa, S.V. and Dommergues Y.R. Eds., Elsevier, Amsterdam. 107pp.

Martin, C. and Torres, H. (1989). Comparative sessitivity of *Rhizoctonia solani* and *Rhizoctonia* like fungi to selected fungicides *in vitro.* . *Phytopathology*. **74**(7): 778-781.

Meah M. B. (2003). Devlopment of an integrated approach for management of phomopsis blight of and fruit rot of eggplant in Bangladesh. Annual research report (2002-2003). 57pp. Department of Plant Pathology, Bangladesh Agricultural University, Mymensingh, Bangadesh.

Meah, M.B. (2007). Formulation of biopesticide in controlling phomopsis rot, foot/collar rot and shoot fruit border of eggplant. Annual research report, USDA-Bangladesh collaborative research. 4-11 p.

Meah, M.B. and Islam, M.M. (2005). Development of an integrated approach for management of Phomopsis blight and fruit rot of eggplant in Bangladesh. Annual Research Report (2004-2005), Department of Plant Pathology, BAU, Mymensingh. p.42.

Meah, M.B., Islam, M.R. and Islam, M.M. (2004). Development of an integrated approach for management of Phomopsis Blight and Fruit rot of Eggplant in Bamgladesh. Ann. Res. Reptr. Dept. of Plant Pathology, BAU, Mymensingh, Bangladesh. p.62.

Mirkova, E. (1982). In vitro study of the antagonistic activity of *Trichoderma* spp. towards some soil pathogens. Gradinarskai. I. Lozarska Nauka. **19**(1): 73-81.

Moity, T.H.A.E. and Shalta, M.N. (1981). Biological control of white rot disease *Sclerotium* sp. of onion by *Trichpderma harzianum*, *Phytopathol. Z.* **100**: 29.

Monaco, C.A., Alippi, H.E. and Pasquare, A.O. (1991). *Trichoderma spp.* a biological agent of *Fusarium spp.* and *Sclerotium rolfsii*. *Adv.Hort. Sci.* **5**(3): 92-95.

Mukhapadhyay, A.N. (1995). Exploitation of *Gliocladium virens* and *Trichoderma harzianum* for biological seed treatment against seed borne disease. *Indian J. Mycol. Plant Pathol.* **2**(1&2): 124.

Mukharjee, PK., Sarmah, D.K. and Shrestha, S.M. (1995). Comperative antagonism properties of *Gliocladium virens* and *Trichoderma harzianum on Sclerotiorum rolfsii and Rhizoctonia solani-its* relevens to understanding the mechanisms of biocontrol. *J. Phytopathol*. **143**(5): 275-279.

Mukhopadhyay, A.N. (1989). National seminar and 7th Workshop of AICRP on Biological Control, Lucknow. Oct. 23-25 pp.

Nene, Y.L.; Sheila, V.K. and Sharma, S.B. (1996). A work list of chickpea and pigeonpea pathogens (5^{th} ed.) ICRISAT, Patanecheru, Andra Pradesh. 27p.

Ortiz, A. and Orduz, S. (2001). In vitro evaluation of *Trichoderma* and *Gliocladium* antagonism against the symbiotic fungus of the leaf-cutting ant Attacephalotes. *Mycopathologia*. **150**: 53–60.

Ozbay, N., Newman, S.E. and Brown, W.M. (2004). Evalution of *Trichoderma harzianum* strains to control crown and root rot of greenhouse fresh market tomatoes. Horticulture and Landscape Architecture, Colorado State University, Fort Collins, Colorado, USA. Acta-Horticulturae. **635**:79-85.

Pandya, J.R., Sabalpara, A.N. and Chawda, S.K. (2011) *Trichoderma*: a particular weapon for biological control of phytopathogens. *J. of Agril. Tech.* **7**(5): 1187-1191.

Papavizas, G. C. (1985). *Trichoderma* and *Gliocladium*: biology, ecology, and potential for biocontrol. *Annu. Rev. Phytopathol*. **3**: 23-54.

Papavizas, G.C. and Lewis, J. A. (1985). Introduction augmentation of microbial antagonists for the soil-borne plant pathogens. In : Biological control in crop production. *Ed. Allenheld, Totowa, N. J.* 305.

Rangasami, G. (1988). Disease of crop plant of India. Printice-Hall of India Private Limited. New Delhi. 101p.

Rettinassababady, C. and Ramadoss, N. (2000). Effect of different substrates on the growth and sporulation of *Trichoderma viride* native isolates. *Agril. Sci. Digest.* **20**(3): 150-152.

Roberti, R., Flori, P. and Pisi, A. (1996). Biological control of soil brone *Sclerotium rolfsii* infection by treatment of bean seeds with species of *Trichoderma*. *Petria.* **6**(2): 105-116.

Sangeetha, P. Jeyarajan, R. and Panicher, S. (1993). Mass multiplication of bio-control agent *Trichoderma spp*. *Indian J. Mycol. Plant Pathol*. **23**(3): 328-330.

Shahiduzzaman, (2009). Formulation of Compost-cum-biopesticide using Municipality waste. MS thesis, Dept. of Plant Pathology. Bangladesh Agricultural University, Mymensingh, Bangladesh. 54p.

Shamarao, J., Siddaramaidah, A.L., Narayanaswamy, H. and Jahagirdar, S. (1998). Screening of substrates of mass multiplication of *Trichoderma viride*. *Karnataka J. Agril. Sci.* **11**(1): 233-236.

Shamsuzzaman., Islam, S.M.A. and Hossain, I. (2003a). Production of *Trichoderma* conidia in agro-waste. *Bangladesh J. Environ. Sci.* **9**: 146-150.

Shamsuzzaman., Islam, S.M.A. and Hossain, I. (2003b). *Trichoderma* culture and germination of sweet gourd seed. *Bangladesh J. Seed Sci. & Tech.* **7**(1&2): 91-95.

Shin, G.C., Im. G.J., Yu, S.H. and Park, J.S. (1987). Biological control of sesame soil-borne disease by antifungal micro-organisms. *Korean J. Plant Protec*. **26** (4): 229-237.

Singh, R.S. (1984). Diseases of vegetable crops. Oxford & IBH Publishing Co. New Delhi. 512p.

Sivan, A. and Chet, I. (1986). Biological control of *Fusarium* spp. in cotton, wheat and muskmelon by *Trichoderma harzianum*. *J. Phytopathol.* **116**(1): 39-47.

Sivan, A., Elad, Y. and Chet, I. (1984). Biological control effects on a new isolate of *Trichoderma harzianum* on *Pythium aphanidermatum*. *Phytopathology.* **74**: 498-501.

Strashnow, Y., Elad, Y., Sivan, A., Rudich, Y. and Chet, I. (1985). Control of *Rhizoctonia solani* fruit rot of tomatoes by *Trichoderma harzianum*. *Crop Protec*. **4**(3): 359-364.

Sugha, S.K. Sharma, B.K. and Tyagi, P.D. (1993). Factors affecting development of collar rot of gram (*Cicer arietinum*) caused by *Sclerotium rolfsii*. *Indian J. Agril. Sci*. **63**(6): 382-385.

Sultana, N. and Hossain, I. (1999). Biological control of foot and root rot of lentil with *Trichoderma harzianum. Bangladesh J. Seed Sci. & Tech.* **3**(1&2): 107-111.

Sultana, N., Chowdhury, M.S.M. and Hossain, I. (2001). Growth and storability of *Trichoderma harzianum* and its effect on germination of tomato seeds. *Bangladesh J. Seed Sci. & Tech*. **5**(1&2): 117-121.

Suseelendra, Desai and E. Schlosser. (1999). Parasitism of *Sclerotium rolfsii* by *Trichoderma*. *Indian Phytopath*. **52**(1): 47-50.

Talukder, M.J. (1974). Plant Diseases in Bangladesh. *Bangladesh J. Agril. Res*. **1**(1):61-68.

Tran, N.Ha. (2010). Using *Trichoderma* species for biological control of plant pathogens in *Viet Nam*. *J. ISSAAS* . **16**(1):17-21.

Tverdyukov, A.P., Nikonov, P.V. and Yushchenko, N.P. (1994). *Trichoderma. Rev. Plant Pathol*. **739**(4): 273.

Waraitch, K.S., Kanawer, R.S., Bipen, K. and Kumar, B. (1986). Fungicidal control of *Sclerotium* root rot of sugar beet (*Beta vulgaris*) caused by *Sclerotium rolfsii*. *Indian Phytopathol*. **39**(1): 100-102.

Wells, H.D., Bell, D.K. and Jaworski, C.A. (1972). Efficacy of *Trichoderma harzianum* as a biocontrol of Sclerotium rolfsii. *Phytopathology*. 62: 442.

Wells, H.D., Bell, D.K. and Jaworski, C.A. (1972). Efficacy of *Trichoderma harzianum* as a biological control for *Sclerotium rolfsii*. *Phytopathology*. **62**(4): 442-447.

Whipps, J.M. and Lumsden, R.D. (2001). Commercial use of fungi as plant disease biological control agents: status and prospects, in: T. Butt, C. Jackson, N. Magan (Eds.), Fungal Biocontrol Agents: Progress, Problems and Potential, CABI Publishing, Wallingford. pp. 9–22.

Xu, T., Zhong, J.P. and Li, D.P. (1993). Antagonism of *Trichoderma harzianum* T82 and *Trichoderma* species NF9 against soil and seed borne pathogens. *Acta. Phytopathol. Ca. Scinica*, **23**(1): 63-67.

APPENDICES

Appendix 1. Composition of Potato Dextrose Agar (PDA) Media:

The compositions of the media used in this thesis work are given below: This media were autoclaved at 121^{O}c for 15 minutes at 15 lb pressure.

Ingredients	**g/L**
Peeled Potato	200g
Dextrose	20g
Agar	17g
Water	1000ml

Appendix 2. Market prices of different substrates:

Prices of different substrates used in this thesis work are given below:

Substrates	Price (Tk/kg)
Rice bran	15
Wheat bran	15
Lentil bran	16
Gram bran	15
Black gram bran	18
Grass pea bran	20
Mustard oil cake	22
Saw dust	15