EVALUATION OF *TRICHODERMA* BASED COMMERCIAL BIO-FUNGICIDES IN CONTROLLING FOOT AND ROOT ROT DISEASE OF LENTIL CAUSED BY *SCLEROTIUM ROLFSII*

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CERTIFICATE

This is to certify that the thesis entitled, "**EVALUATION OF *TRICHODERMA* BASED COMMERCIAL BIO-FUNGICIDES IN CONTROLLING FOOT AND ROOT ROT DISEASE OF LENTIL CAUSED BY *SCLEROTIUM ROLFSII***" *submitted to the Faculty of Agriculture, Sher-e-Bangla Agricultural University, Dhaka, in partial fulfillment of the requirements for the degree of* ***MASTER OF SCIENCE*** *in* ***PLANT PATHOLOGY****, embodies the result of a piece of bona fide research work carried out by* ***SURAIYA AKHTAR RAKHI****,* ***Registration No. 18-09139*** *under my supervision and guidance. No part of the thesis has been submitted for any other degree or diploma.*

I further certify that such help or source of information, as has been availed of during the course of this investigation has duly been acknowledged.

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LIST OF ABBREVIATIONS

ABBREVIATIONS	FULL WORD
%	Percentage
PDA	Potato Dextrose Agar
G	Gram
^{o}C	Degree celcius
Psi	Per square inch
Cm	Centimeter
et al.	and others (at ell)
CRD	Complete Randomized Design
RCBD	Randomized Complete Block Design
DAI	Days After Inoculation
DAS	Days After Sowing
sp.	Species
etc.	Et cetera
Viz.	Videlicet (namely)

EVALUATION OF *TRICHODERMA* BASED COMMERCIAL BIO-FUNGICIDES IN CONTROLLING FOOT AND ROOT ROT DISEASE OF LENTIL CAUSED BY *SCLEROTIUM ROLFSII*

ABSTRACT

Four experiments were conducted to evaluate commercial bio-fungicides for the management of foot and root rot disease of lentil at Sher-e-Bangla Agricultural University, Dhaka, Bangladesh during November 2019 to April 2020 following CRD and RCBD design with 3 replications for *in vitro* and *in vivo* experiments, respectively. Susceptible variety BARI Masur -1 (Utfala) was used in this experiment. Nine *Trichoderma* based commercial bio-fungicides viz. G-derma (powder), Bio-derma (powder), Decoprima, Recharge, Bio-derma (peat soil), G-derma (LDS), Terrabio, Decohumate, Ttricost and two chemicals viz. Formaldehyde and Autostin were assessed in this study. In pot experiment, seed treatment with bio-fungicides was done and the lowest disease incidence (6.48%) was recorded from T_5 (Tricost) and the highest disease incidence (54.91%) was observed from T_0 (Control). Significantly the highest yield /pot (12.55 g) was found in T_5 (Tricost) treatment. In another pot experiment, soil treatment with bio-fungicides was done and the lowest disease incidence (4.84%) was recorded from T_5 (Bio-derma - peat soil) and the highest disease incidence (53.29%) was observed from T_0 (Control). Significantly the highest yield /pot (16.83 g) was found in T_5 (Bio-derma- Peat soil) treatment. Moreover, in case of seed and soil treatment with bio-fungicides, the lowest disease incidence (4.87%) was recorded from T_4 (Decoprima) and the highest disease incidence (49.92%) was observed from T_0 (control). Significantly the highest yield /pot (17.83 g) was found in T_4 (Decoprima) treatment. In field experiment, the lowest disease incidence (10%) was recorded from T_5 (Tricost) and the highest disease incidence (48.89%) was observed from T_0 (Control). The highest yield /plot (437.67 g) and yield/m^2 (145.89g) found in T_5 (Tricost) treatment. Thus, seed and soil treatment with *Trichoderma* based bio-fungicides significantly reduced foot and root rot disease of lentil.

CHAPTER 1

INTRODUCTION

Lentil is one of the early domesticated plant species, as old as those of einkorn, emmer, barley and pea (Harlan, 1992). Lentil *(Lens culinaris*) belongs to the family Leguminosae is an important pulse crop in semiarid regions of Iran, India, Turkey and Canada and originated in the Fertile Crescent of the Near East, and dates back to the beginning of agriculture itself (Sabagh pour *et al.*, 2004). Globally, it is cultivated as a rainfed crop on 3.85 million hectares (m ha) area with 3.59 million ton (mt) production (Erskine *et al.*, 2011). Canada, India, Turkey, Australia, United States of America, Nepal, China, and Ethiopia are the major players in global lentil production (FAO, 2013). Both red and green lentils are produced in the region with variable proportion. It is an important crop in food, feed and farming systems of West Asia and North and East Africa (Akibode, 2011). It has been established in a wide range of agro-ecology but production is limited to tropical areas. The spread of lentil from the center of origin has been accompanied by the selection of traits important for adaptation to environments that can be climate, soil and their impact on season length, abiotic and biotic stresses (Materne *et al*., 2009). Lentil is currently an important pulse crop grown widely throughout the Indian Subcontinent, Middle East, Northern Africa and East Africa, Southern Europe, North and South America, Australia and West Asia (Erskine, 1997 and Taylor, 2003). It is a primary component for farming systems of those areas (Sarker, 2011). The major lentil-growing countries of the world are Canada, India, Turkey,

Australia, USA, Nepal, China, and Ethiopia. Out of the total increased volume of global production in recent years, the most is coming mainly from Canada and India. The total lentil cultivated area in the world is estimated around 4.34 million hectares with annual production and productivity of 4.95 million tons and 1260 $Kgha^{-1}$ respectively (FAO, 2014). The production of the crop is increased significantly from year to year through expansion of net cropped area along with its productivity. This increment was stimulated by greater improvement in demand of both domestic and international market of the crop. Even though lentil producing nations are striding in skyrocketing of their production to fill domestic demand and overwhelming of the export market but still the supply gap remains wide which is aggravated by rapid population growth, ever changing client demand and limitation in genetic improvement of the crop. Most of the production which reaches around 56% is consumed locally and only 44% of the production is supply to the global market (Kumar *et al*., 2013). Canada is the Leading exporting nation, while India is the leading lentil consuming and producing nation (Bedard *et al*., 2009). The demands for lentil in these two regions are expected to rise further due to population growth and rising income. By 2030, the world lentil consumption is estimated at 5.5 mt, being an increase of almost 2 mt from the present level (Clancey, 2009).

Lentil, a member of the legume family, Leguminosae which is capable of fixing and utilizing atmospheric nitrogen through symbiotic relationship with Rhizobium at the root nodule of the crop. It provides a good source of protein (20% to 30%), but are limited in the amino acids methionine and cysteine (Kandel and Ashley, 2013) and 48% carbohydrate (Feedipedia, 2012). Lentil is also used for human consumption as a protein source in a

diverse range of product and is an excellent source of vitamin A and provides fiber, potassium, B vitamins, and iron (Kochhar, 2009).

Pulses are important legume crops in Bangladesh because of their importance in food, feed, and cropping systems. It contains about twice as much protein as cereals. Pulses have played an important role in sustaining the productivity of soils in Bangladesh for centuries. They are generally grown without fertilizer since they can meet their nitrogen requirement by symbiotic fixation of atmospheric nitrogen in the soil (Senanayake *et al.*, 1987; Zapata *et al.*, 1987; Fried and Middleboe, 1977). The per capital consumption of pulse in Bangladesh is only 12 g/day, which is much lower than WHO recommendation of 45 g/day (Afzal *et al.*, 1999). In Bangladesh, lentil placed second position among the pulses according to area and production but stand first in terms of usage (Afzal *et al.*, 1999). It is the principal and popular edible crop among pulses. The area, production, and yield of lentil in Bangladesh were 208800 ha, 153000 tons (t), and 0.733 t/ha, respectively, in 1991-92 (BBS, 1995). After 18 years, the area, production, and yield of lentil were 70983 ha, 60537 t, and 0.853 t/ha, respectively, in 2008-09 (BBS, 2009). Thus, it is noted that area and production of lentil decreased 2.94 and 2.53 times, respectively. Total production of lentil in Bangladesh is about 164,000 metric tons (BBS, 2017).

It is specially honored as a protein source in comparison with high cost animal protein and considered as poor man's meat (Begum, 1997). In Bangladesh lentil is cultivated in an area of 1,54,655 ha and the yearly production is about 1,22,000 mt (BBS 2009). However, the yield of lentil is much lower in Bangladesh compared to that of other lentil growing countries

like Syria, Turkey, Canada, USA and Ethiopia (Hossain *et al*., 1999). It is a cheap source of protein for human beings and also for animals in country (Sattar *et al*., 1996). As the price of animal protein is increasing day by day, the protein storage in the diet system of the people in the country can be met up through improvement and increasing the production of lentil.

The lower yield quality of lentil is because of poor management practices, scarcity of good quality seeds, delayed sowing, low yield potential of local cultivars and improper plant disease management. Diseases play an important role in yield reduction in crop plants. Productivity of lentil is reduced by pathogens through infection and damage to leaves, stems, roots and pods. It also reduces marketability due to discoloration of the leaves. It causes seedling death at early stage resulting very poor plant stand which ultimately produces very low yield.

Lentil is affected by wide range of diseases like fungal, bacterial etc. The most dangerous enemy of lentil (*Lens culinaris*) plant is fungus (BARI, 2005). Lentil is affected by wide range of fungal diseases (Agrawal, 1979). The productivity of lentil reduces because of infection through roots and collars of the plant. The market value of products is hampered due to the discoloration of seeds.

Lentil suffer from attack of a number seed and soil borne diseases such as vascular wilt, collar rot, root rot, stem rot, rust, powdery mildew and downy mildew, which are caused by *Fusarium oxysporum* f. sp. *lentis*, *Sclerotium rolfsii*, *Rhizoctonia solani*, *Uromycis fabae*, *Erysiphe polygoni* and

Peronospora lentis, respectively (Singh and Tripathy, 1999; Khare *et al*., 1979).

In Bangladesh, among the diseases of lentil, foot and root rot disease caused by *Sclerotium rolfsii* is a potential threat to lentil production. Bakr (1994) observed 84% loss in pulse due to infection of *Sclerotium rolfsii*. It is a non-specialized soil borne fungal pathogen of the world and it has wide host range of over 500 crop species (Mehrotra and Aneja, 1990). In Bangladesh, about 44% lentil plants are infected by foot and root rot disease (Anonymous, 1986). This disease affects mainly in roots, leading to poor emergence of seedlings, stunted growth of plants and reduced yields. Symptoms include sunken lesion and brown or black discoloration on roots, shrinking root system and root decay. If they develop at all, nodules are less numerous, smaller and pale in color. In plants growing from infected seeds, seedlings can blight shortly after emergence. The plants that survive are chlorotic and have poor vigor. Plants infected during late stages of development show stunted growth. Opportunistic pathogens colonize and feed on decaying tissue which makes the symptoms worse. In the field condition the disease often occurs in patches and may expand if conditions are favorable for the pathogens. Foot and root rot caused by *Fusarium oxysporum* and *Sclerotium rolfsii* is considered as an important and destructive disease of pulses in almost all legume-growing countries of the world including Bangladesh (Anon., 1986, Dey *et al*., 1993). In Bangladesh, about 44% lentil plants are infected by foot and root rot disease (Anon., 1986). It causes seedling death at early stage resulting very poor plant stand which ultimately produces very low yield.

Being soil borne pathogen, *S. rolfsii* is difficult to control. High soil moisture and high temperature accelerate disease development and under favorable environmental condition incidence of foot and root rot of lentil goes up to 80-90% (Grichar and Bosweel, 1987). Wever (1931) and Garret (1956) reported that the fungus survived in the soil for years together by producing sclerotial bodies and causes the disease on various hosts. For the potential control of the sclerotia forming pathogen single option may not be effective. Integrated disease management may be effective to control this disease. Moreover, indiscriminate use of chemical and fertilizer in agriculture has resulted in the development of several problems such as pesticide resistance in pests and pathogens, resurgence in target and non-target pests, destruction of beneficial organisms like honey bees, and chemical residue in food, feed and fodder.

The biological management of soil-borne diseases is increasingly gaining stature as a possible practical and safe approach. The potential of the antagonistic micro-organisms in reducing the intensity of crop damage by the soil-borne plant pathogens has been reported (Lewis and Larkin, 1997). Several strains of *Trichoderma* spp. have been found to be effective as bio-control agents of various soil-borne plant pathogenic fungi such as *Fusarium, Pythium, Rhizoctonia* and *Sclerotium* (Papavizas, 1985). *Trichoderma* spp. is found in almost any tropical and temperate soil. The suppression of disease by *Trichoderma* is based on hyper parasitism, antibiosis, induced resistance in the host plant and competition for nutrients and space (Harman *et al*., 2004). Bio-control agent like *Trichoderma harzianum* is reported to have great effect against soil borne pathogen (Moon *et al.*, 1988; Singh *et al.*, 1997). Successful bio-control against *S.*

rolfsii using *Trichoderma* spp. has been reported by many researchers (Elad *et al*., 1980; Sreennivasaprasad and Manibhushanrao, 1993; Dey *et al*., 2004).

Bio-sourced fungicides refer to the direct use of biologically active substances produced by biological organisms or biological metabolic processes or substances extracted from organisms as disease prevention and treatment. The concept of biological pesticides is vague and can be considered equivalent to biologically derived fungicides. According to statistics, there are more than 100 kinds of biological pesticide products in the world, but more than 90% are bio-insecticides. The next important commercial bio-pesticide will be bio fungicide products those have strong selectivity and safety for humans and animals. At present, most of the bio fungicides developed and widely applied in the market have effects only on diseases, such as humans, animals, and various beneficial organisms (including natural enemies of animals, natural enemies of insects, bees, pollinators and aquatic organisms such as fish and shrimp). It is safer and has less impact on non-target organisms. The biological fungicide mainly uses the special disease prevention and growth-promoting function of certain special microorganisms or metabolites of microorganisms to achieve the control effect. Its effective active ingredients are completely present and derived from natural ecosystems.

At present, the main means of controlling plant diseases are chemical fungicides. However, due to potential human health hazards, environmental pollution, non-target organisms, and the development of plant-pathogen resistance, the development of chemical fungicide has been limited.

Moreover, control of soil-borne pathogen is difficult by using chemical fungicides. Thus, biological fungicides are attracting more and more people's attention and interest. Considering above facts, this research work is designed to achieve the following objectives:

- To detect and identify of the causal organisms of foot and root rot disease of lentil;
- To evaluate seed treatment by commercial bio-fungicides against foot and root rot disease of lentil;
- To evaluate soil treatment by commercial bio-fungicides against foot and root rot disease of lentil; and
- To evaluate seed and soil treatment by commercial bio-fungicides against foot and root rot disease of lentil.

CHAPTER 2
REVIEW OF LITERATURE

Foot and root rot disease of lentil caused by *Fusarium oxysporum* and *Sclerotium rolfsii* is a common and most important disease in our country. This disease causes serious yield loss of the crop. Researchers all over the world have carried out intensive investigation on the foot & root rot of lentil. Literature in relation to management, disease incidence and yield loss assessment of foot & root rot disease of lentil is reviewed and presented in this chapter.

2.1. Foot and root rot disease of lentil

Eliane (2017) stated that root rot is a serious threat to agriculture worldwide, continuously reducing yields and crop survival. Depending on the causal agent, host susceptibility, and the environmental conditions, entire fields can be lost to this disease. Oomycetes and fungi have been found to be the most commonly widespread root rot pathogens. Several fungi were reported to cause root rot disease, including, *Rhizoctonia* spp., *Fusarium* spp. *and Phoma* spp., *Aphanomyces euteiches* and *Thielaviopsis basicola*. These diseases are highly influenced by the environment with a broad range of hosts, hidden underground symptoms and overwinter structures of many root rot pathogens, disease control and management are very complex and hard to achieve. Overall, despite the complexity of this trait, resistance to root rot through enhanced varieties is the biggest promise to control such devastating diseases.

Begum and Bhuiyan (2007) stated that, among the major diseases of lentil foot and root rot is the most important one. The disease is caused by a soil born fungus and it may cause 100% seedling mortality in the field under monoculture and conducive weather condition.

Bakr *et al*., (2007) reported that, foot rot disease of lentil cause up to 44.40% yield reduction in Bangladesh.

Anonymous (1986) reported that, foot and root rot caused by *Fusarium oxysporum* and *Sclerotium rolfsii* is considered as an important and destructive disease of pulses in almost all legume-growing countries of the world including Bangladesh. In Bangladesh, about 44% lentil plants are infected by foot and root rot disease. It causes seedling death at early stage resulting very poor plant stand which ultimately produces very low yield.

Ahmed (1985) and Bakr (1986) reported that, fifteen pathogens causing 17 diseases in lentil crop have so far been recorded in Bangladesh but only few are severe causing losses in yield. These are rust (*Uromyces fabae*), stemphylium blight (*Stemphylium* sp), wilt *(Fusarium oxysporum*) and foot and root rot (*Sclerotium rolfsii*).

2.2. Symptoms of foot and root rot disease of lentil

Eliane (2017) stated that, root rot symptoms are a major threat because the damage starts below the ground, where the first symptoms are not discernible. When the symptoms become apparent on the above ground part of the plant, yield is already compromised and plant survival is jeopardized.

Weidong *et al*., (2011) reported that patches are found in the field in case of foot and root rot disease of lentil. Top leaves in the plant may wilt and droop. They can also shrink and curl without defoliating early. Some other symptoms include yellowing, reduced root system with discoloration, poorly developed nodules, and damage at the taproot tip. Seeds may rot, pre-emergence damping off can occur, and the plant may die.

According to Begum and Bhuiyan (2007), among various diseases of lentil, foot and root rot is the important one. It may cause 100% morality of seedlings in the field under monoculture and conductive weather condition.

Bakr (1986) stated that, foot rot is mainly a seedling disease attacking the crop usually up to 30 days of germination. The fungal stands along with mustard seed like sclerotia are generally observed associated with infected portion at soil level. He also explained that the top roots are infected and normal growth is arrested which may give stunted appearance and finally causing wilting and dying of plants. It may also observe brown discoloration involving pith and xylem.

2.3. Causal organism of foot and root rot

Singh and Tripathy (1999) stated that, lentil suffer from attack of a number seed and soil borne diseases such as vascular wilt, collar rot, root rot, stem rot, rust, powdery mildew and downy mildew, which are caused by *Fusarium oxysporum* f. sp. *lentis*, *Sclerotium rolfsii*, *Rhizoctonia solani*, *Uromycis fabae, Erysiphe polygoni* and *Peronospora lentis*, respectively . In Bangladesh, among the diseases of lentil, foot and root rot disease caused by *Sclerotium rolfsii* is a potential threat to lentil production.

Dey et al., (1993) reported that, foot and root rot caused by *Fusarium oxysporum* and *Sclerotium rolfsii* considered as an important and destructive disease of pulses in almost all legume-growing countries of the world including Bangladesh. In Bangladesh, about 44% lentil plants are infected by foot and root rot disease. It causes seedling death at early stage resulting very poor plant stand which ultimately produces very low yield (Anon., 1986).

Punja *et al.*, (1985) reported that, sclerotia of *Sclerotium rolfsii* are produced a large number or on adjacent to infected plant tissue and can survive in soil for 1-3 years. They are capable of initiating infection with or without a food base of organic matters.

Maiti and Sen (1984) reported that, sclerotia had a 91% survival rate after 222 days in natural soil at 50% water holding capacity.

Katti *et al.*, (1983) found that, maximum survival rate of fungus at 30-50% soil moisture and at temperature between 20-25°C.

Ahmed (1980) stated that, *Sclerotium rolfsii* is facultative saprophyte and can maintain continuity of generation under adverse condition by the formation of brown sclerotia. *Sclerotium rolfsii* is a soil borne pathogen reported by Sumbali and Mehrotra (1980).

Treggi (1956) found that, *Sclerotium rolfsii* grew vigorously at temperature between 30- 33 °C.

2.4. Management of Foot and root rot disease of lentil by commercial bio-fungicides

Jahan *et al*., (2017) stated that, efficacy of IPM Lab bio-pesticide for controlling Sclerotium rot of sugarbeet was studied. *Sclerotium rolfsii* was identified as causal organism of Sclerotium rot of sugarbeet. In an *in-vitro* assay it was observed that the growth of *S. rolfsii* was inhibited by *Trichoderma harzianum* based IPM lab bio-pesticide indicating antagonistic effect against *S. rolfsii*. From the pot experiment, it was observed that plants treated with bio-pesticide (*Trichoderma* based) reduced Sclerotium rot infection. A field experiment was conducted to investigate the efficacy of *Trichoderma* based bio-pesticide to control Sclerotium rot of sugarbeet. After germination of sugarbeet plants, periodically infections were recorded and results showed that incidence of Sclerotium rot of sugarbeet was very low in bio-pesticide treated plots, where the best performance was recorded with bio-pesticide treatment at 80 kg/ha. The highest dose (80 kg/ha) of bio-pesticide used in this experiment showed significantly better growth, the highest yield (65.0 t/ha) and lower Sclerotium rot incidence (10.67%) than the non-treated plots. Thus, *Trichoderma* based IPM Lab biopesticide is effective in reducing Sclerotium rot of sugarbeet.

Dwivedi and Ganesh (2016) reported that, *Sclerotium rolfsii* is soil-borne saprophytic fungus which causes different types of diseases like collar-rot, sclerotium wilt, stem-rot, charcoal rot, seedling blight, damping-off, foot-rot,

stem blight and root-rot in more than 500 plants species including tomato, chilli, sunflower, cucumber, brinjal, soybean, maize, groundnut, bean, watermelon etc. *S. rolfsii* may be controlled through biological agents (*Trichoderma harzianum, T .viride T. asperellum, Penicillium* sp, *Curvularia* sp, *Aspergillus niger, Bacillus subtilis, Pseudomonas fluorescens, Pseudomonas cf.montelii, P. aeruginosa*. Among all these control measures, biological, soil solarization and medicinal plant extract were the more significant than chemical control.

Khalequzzaman (2016) conducted an experiment at the sick plot, Pulses Research Centre, Ishurdi, Pabna, Bangladesh to find out the effect of chemical, botanicals, bio-control agents & healthy seeds against foot & root rot of lentil. The lowest foot & root rot (21.67%) was obtained from seed treatment with Provax 200 (2.5 g/kg seed) followed by seed treatment with *Trichoderma harzianum* compost (1:5) and apparently healthy seeds, while the highest incidence (41.5%) was obtained from untreated control. The highest number of pod/plant (45.26), number of seeds/plant (87.80), weight of 100 seeds/ plant (2.44 g) and yield (1845 kg/ha) were recorded in case of seed treatment with Provax 200 (2.5 g/kg seed) which were followed by seed treatment with *Trichoderma harzianum* compost (1:5) and apparently healthy seeds .

Pratibha *et al*., (2016) reported that, stem and root rot complex disease of groundnut is a devastating disease caused by *Sclerotium rolfsii* and *Macrophomina phaseolina*. To avoid extensive use of pesticides, biological formulation developed from *Trichoderma harzianum* (Th3) was used as an environment friendly option. In the dual culture method, *T. harzianum* Th3

showed around 62% inhibition against *S. rolfsii* and 71.9% inhibition of *M. phaseolina*. Seed treatment, soil application and drenching with *T. harzianum* Th3 showed minimum disease incidence of 21.6% and 12.6%. Thus it reduces the disease incidence by 66.3 to 78.1% as compared with control. In the same way, there was significant response of application of *T. harzianum* Th3 was observed on plant growth promoting parameters at field level to increase number of pods per plant and shelling % and thereby produces maximum yield of 3.37 and 3.6 t/ha respectively.

Pratibha *et al*., (2014) reported that, species of *Trichoderma* are diverse fungal microbial community known and explored worldwide for their versatilities as bio-control and growth promoting agents. They are also widely exploited in industries as sources of enzymes. A large number of research groups are working on various aspects of *Trichoderma* viz., diversity, ecology and their applications. In India, about 110 groups representing various universities and research institutes are working with about 15 different species and have published about 460 research papers. *Trichoderma harzianum* and *Trichoderma viride* are the widely used species and have been exploited on about 87 different crops and about 70 soil-borne and 18 foliar pathogens, respectively. This review aims to give an overview of the status of usage of *Trichoderma* on important agricultural crops by different groups and organizations in the country

Hoque *et al*., (2015) stated that, an investigation was carried out to evaluate the six selected isolates of three bio-control agents against foot and root rot pathogens. The pathogens, *Fusarium oxysporum* and *Sclerotium rolfsii* were isolated from foot and root rot infected lentil seedlings. Four isolates of

Rhizobium leguminosarum, one isolate of *Pseudomonas fluorescens* and one isolate of *Trichoderma harzianum* were used as bio-control agents. In dual culture method, highest zone of inhibition of *F. oxysporum* (57.37%) was measured against *R. leguminosarum* isolate 3 and isolate 4. In case of *S. rolfsii*, 80% and 37.85% inhibition zone were measured against *P. fluorescens* and *T. harzianum*, respectively. In paper towel and water agar test tube tests, seed treatment with all the bio-control agents showed significantly better germination than control. It also resulted in significant increase in shoot and root length that gave high vigor index. In paper towel test, minimum number of deed seeds (9.00), no abnormal and diseased seedlings were counted from *R. leguminosarum* treated seeds. In water agar test tube test, minimum number of deed seed (12.00) and abnormal seedlings (2.00) were counted from *R. leguminosarum* treated seeds. Here, no diseased seedling was found from *T. harzianum* treated seeds.

Shahiduzzaman (2015) observed that, seed treatment with all the tested fungicides/ botanicals reduced the disease severity and increased pod number and crop yield of lentil as compared to untreated control. Provax 200, Bavistin 50WP and *Trichoderma* compost showed better performance than other treatments in both the seasons. However, efficacy of fungicides and botanicals in controlling 715 in 2011-12 cropping season, Bavistin 50WP, *Trichoderma* compost and Neem leaf extract showed statistically identical mortality, pods per plant and yield while in 2012-13 cropping season, Bavistin 50WP and *Trichoderma* compost showed statistically similar results.

Sultana *et al*., (2015) carried out an experiment to evaluate six selected isolates of three bio-control agents against foot and root rot pathogens. The pathogens, *Fusarium oxysporum* and *Sclerotium rolfsii* were isolated from foot & root rot infected seedlings of lentils. Four isolates of *Rhizobium leguminosarum*, one isolate of *Pseudomonas fluorescens* and one isolate of *Trichoderma harzianum* were used as bio-control agents. Using dual culture method, the highest zone of inhibition of *F. oxysporum* (57.37%) was recorded against *R. leguminosarum* isolate 3 & isolate 4. In case of *S. rolfsii*, 80% & 37.85% inhibition zone were measured against *P. fluorescens* and T. *harzianum*, respectively. In paper towel & water agar test tube tests, minimum number of deed seeds (9), no abnormal & infected seedlings were counted from *R. leguminosarum* applied seeds. In water agar test tube test, lowest number of deed seed (12) & abnormal seedlings (2) were counted from *R. leguminosarum* incorporated seeds. No diseased seedling was found from *T. harzianum* treated seeds.

Faruq *et al*., (2014) stated that, efficacy of eight different carrier materials and their combinations were tested to formulate a suitable *Trichoderma harzianum* based bio-fungicides for controlling foot and root rot diseases of brinjal caused by *Sclerotium rolfsii* in tray soil as well as seed bed soil under net house condition of Bangladesh Agricultural Research Institute (BARI). The results from a series of experiments revealed that four combination of carrier materials based *T. harzianum* bio-fungicides such as (1) wheat bran + rice bran, (2) wheat bran + mustard oil cake (MOC) + rice bran, (3) khesari bran + rice bran, and (4) khesari bran +MOC+ rice bran were suitable for controlling the soil borne foot and root rot disease (*S. rolfsii*) of brinjal in tray soil as well as seed bed soil conditions.

Kashem *et al*., (2014) reported that, ten bio-agents including control were tested in these experiments. Macerated extract of *Fusarium solani* + *Trichoderma harzianum* showed the best result in controlling root rot of lentil with the highest seed germination (100%), radical length (1.56 cm), seedling emergence (95.73%), root length (8.16 cm), shoot length (17.75 cm), number of branches/5 plants (15.56). The lowest root rot (25.93%) was also observed in the treatment of *Fusarium solani* + *Trichoderma harzianum*. The other four treatments, macerated extract of *Sclerotium rolfsii*, *Fusarium solani*, *Trichoderma harzianum, Sclerotium rolfsii* + *Trichoderma harzianum* showed better results over control in terms of all plant parameters.

Saigan *et al*., (2008) conducted an experiment and the aim of that two years research work was to recognize bio control agents and its antagonistic efficiency on *Sclerotium rolfsii* in vitro condition. *Sclerotium rolfsii* Sacc. is a causal agent of white foot and root rot disease in many plants. This fungi causing damage in tea nurseries of tea cultivated countries especially in Iran. Due to importance of this disease in tea nurseries of Iran and impossibility using of chemical control against this damaging agent. In this research, five species of *Trichoderma viride, Trichoderma harzianum, Trichoderma hamatum, Trichoderma longibrachiatum* and *Trichoderma parceramosum* as antagonist fungi against *Sclerotium rolfsii* were collected and identified from tea nurseries and seedlings in Iran. Then efficiency of these bio control agent against *Sclerotium rolfsii* were investigated *in vitro* condition. Results showed that these species with different mechanism such as lysis of sclerotia, inhibited mycelial growth of *Sclerotium rolfsii* with volatile

metabolites producing and parasitized the hyphal trends of disease agent were showed its antagonistic effects against causal agent of white foot and root rot in tea seedling. *Trichoderma viride, Trichoderma harzianum and Trichoderma hamatum* after 30 days destructed and lysis the sclerotia 98.5, 86.5 and 85%, respectively. Producing of volatile metabolites after 72 h reciprocal growth of *Trichoderma viride, Trichoderma harzianum and Trichoderma longibrachiatum* till 60.8, 54.8 and 54.4% prevented mycelium growth of *Sclerotium rolfsii*, respectively.

CHAPTER 3
METHODS AND MATERIALS

The methods followed and materials that were used in the present research work were stated in this chapter. The experimental site, weather, land preparation, experimental design, lay out, inoculation, growth parameters, data collection were described in the chapter.

3.1. Experimental site

The pot experiments were conducted in the Net House of the Department of Plant Pathology, Sher-e-Bangla Agricultural University, Dhaka-1207. The field experiment was conducted in the field of Central Farm of Sher-e-Bangla Agricultural University, Dhaka-1207. The location for the experimentation site was 23°75N latitude and 90°35E longitude with an elevation of 8.3 meter from sea level.

3.2. Experimental period

The both pot and field experiments were conducted during the *Rabi* season from November 2019 to March 2020. Laboratory research was done from July 2019 to May 2020.

3.3. Soil type

The area of the experimental site was in the sub-tropical zone. The soil of experimental site belongs to the agro-ecological regions of “Madhupur Tract” under AEZ No.28. The top soil of the region is clay loam in texture and olive gray with common fine to medium distinct black yellow brown

mottles. The pH of the soil was 4.47 to 5.55 and organic carbon contents is 0.82 (Appendix I).

3.4. Weather

The monthly mean for daily maximum, minimum and average temperature, relative humidity (RH%), monthly total rainfall and sunshine hours received at the experimental field during the period of the experiment have been collected from Bangladesh Meteorological Department, Agargaon, Dhaka (Appendix II).

3.5. Planting materials

The lentil (*Lens culinaris*) variety BARI Masur - 1 (Utfala) released from Bangladesh Agricultural Research Institute, Joydebpur, Gazipur was used for the experiment. This variety is susceptible to foot and root rot disease. Seeds were collected from Pulses Wing, Bangladesh Agricultural Research Institute, (BARI), Joydebpur, Gazipur, Bangladesh.

3.6. Soil preparation

For pot experiment, the soil was collected from the uppermost 0-15 cm of soil from the experimental field of Sher-e-Bangla Agricultural University, Dhaka. For field experiment, the experimental land had been properly ploughed and cross ploughed and cleaned before seed sowing and application of fertilizers and manure was done in the field. The experimental land was good tilth. Finally, the field was properly leveled before seed sowing. The soil mixed with recommended fertilizers and manures mentioned in Table 1.

3.7. Application of manures and fertilizers

According to the standard recommendations, manures and fertilizers were applied. The following doses were used for carrying out the field experiment suggested by (*Krishi Projukti Hatboi*, 2015).

Table 1. Manures and fertilizers applied in the experiments

Name of Fertilizer	**Rates (kg/ha)**
Urea	40-45
TSP	80-90
MoP	30-40
Zinc sulphate	5
Boric acid	7.5
Cowdung	5000

3.8. Seed sowing

In pot experiments, 60 seeds were sown in each pot. Within 8-10 days, seeds were germinated. For field experiment, 20g seeds were sown in line sowing continuously in the field. The seeds were planted at about 4 cm depth in the soil.

3.9. Intercultural operation

Intercultural operations like weeding, thinning, mulching, irrigation etc. were done specifically in the plots. First irrigation was done immediately after seed sowing. After germination, the irrigation was done for several times at 7 to 15 days intervals. Proper drainage system was maintained to release excess water created by rainfall immediately after stagnation.

Plate 1. Seed sowing in the field

3.9.1. Irrigation

In this experiment, first irrigation was done immediately after seed sowing. After germination irrigation was done several times at 7 days interval by sprinkler. Proper drainage system was maintained both pot and field experiments.

3.9.2. Thinning

Thinning was done after 15 days of sowing and proper distance was maintained.

3.9.3. Weeding

After 20 days of sowing first weeding was done. 2nd weeding was done at 35 days of sowing.

3.10. Treatments

All together 11 treatments were used in pot and field experiments for seed and soil treatments including 9 commercial formulations of *Trichoderma* based bio-fungicides and 2 chemicals viz. formaldehyde and Autostin for comparison.

3.10.1. Collection of treatments

Bio-derma (Powder) and Bio-derma (Peat soil) were collected from Ispahani Agro Ltd., Bangladesh. G-derma (Powder) and G-derma (LDS) were collected from GME Agro Ltd., Bangladesh. Recharge (Powder) was collected from Russell IPM, UK. Decoprima (Powder) was collected from Mohsin Enterprize. Terrabio (Powder) and Decohumat were collected from PT Prima Agro Tech., Indonesia. Tricost was collected from Haychem (Bangladesh) Limited. Formaldehyde (Merck, Germany) was collected from Scientific Market, Tikatuli, Dhaka. Autostin 50 WDG was collected from Auto Crop Care Ltd., Bangladesh.

Tablae 2. Details of the treatments including commercial *Trichoderma* based bio-fungicides formulations

Trade Name/ Commercial Name	Active Ingredients	Formulation	Company
G-derma	*Trichoderma*	Powder	GME Agro Ltd, Bangladesh
Bio- derma	*Trichoderma*	Powder	Ispahani agro Ltd, Bangladesh
Decoprima	*Trichodermma* sp. 4.35×10^5 cfu/g, *Geobacillus* sp. 1.94×10^6 cfu/g *Streptomyces* sp. 1.16×10^6 cfu/g	Powder	Mohsin Enterprize, Bangladesh
Recharge	*Trichoderma*	Powder	Russell IPM, UK
Bio-derma	*Trichoderma*	Peat soil	Ispahani Agro Ltd, Bangladesh
G-derma (LDS)	*Trichoderma*	Liquid	GME Agro Ltd, Bangladesh
Terrabio	*Trichoderma* sp. 4.86×10^6 *Pseudomonas* sp. 1.7×10^7 *Bacillus* sp. 1.93×10^7	Powder	PT Prima Agro Tech. Indonesia
Decohumate	*Trichoderma* 1.5×106 cfu/g	Powder	PT Prima Agro Tech. Indonesia
Tricost	*Trichoderma* 2×106 cfu/g	Powder	Haychem (Bangladesh) Limited
Autostin	Carbendazim	Powder	Auto Crop Care Ltd, Bangladesh
Formaldehyde	37% Formalin	Liquid	Merck, Germany

Bio-derma (powder)

Bio-derma (peat soil)

G-derma (powder)

G-derma (liquid)

Recharge

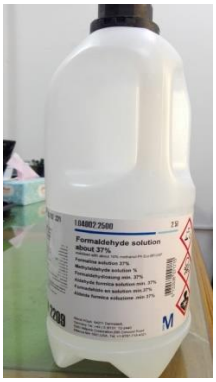
Dicoprima

Terrabio

Decohumat

Tricost

Autostin



Formaldehyde

Plate 2. Different commercial bio-fungicides used for pot and field experiments

3.11. Tagging of plants

In pot experiment, ten plants per pot were maintained for each treatment combinations including control. Randomly three plants were selected from each pot tagged for data collection. Mean values determined to get rating score of each treatment. In field experiment, randomly 10 plants were selected from each plot tagged for data collection.

3.12. Data collection

The data were collected on the following parameters in pot and field experiments:

- Disease incidence (%)
- Germination (%)
- Plant height (cm)
- Number of branch
- Root length (cm)
- Shoot length (cm)
- Vigor index
- Number of pod/plant
- Seed yield/pot (g)
- Seed yield/plant (g)
- Seed yield/ plot (g)
- Seed yield/m^2 (g)

3.13. Procedure of data collection

3.13.1. Disease incidence (%)

Visual observation was done to count total number of diseased/infected plants at different stages of growth. The data on counting total number of diseased plants per pots were recorded at seedling stage 10 days after inoculation (DAI) and consequently at 20 DAI, 30 DAI and 60 DAI. In field experiment, 30 plants were randomly selected and tagged. Then data was collected at 10 days after sowing (DAS), 20 DAS, 30 DAS and 60 DAS. Percent plant diseased per plot was calculated by using the following formula (Agrios, 2005).

$$\text{Disease Incidence (\%)} = \frac{\text{Number of infected plants}}{\text{Number of inspected plants}} \times 100$$

Plate 3: Diseased plants in pot (left) and field (right)

3.13.2. Germination (%)

In pot experiment the germinated seeds were counted. Germination percentage was calculated by using the following formula (ISTA, 1999).

$$\text{Germination (\%)} = \frac{\text{Number of germinated seeds}}{\text{Total number of seeds}} \times 100$$

3.13.3. Plant Height (cm)

Plant height was measured in centimeter by using a meter scale at 30 days after sowing (DAS), 60 and 105 DAS and there after the average of three plants per plot were selected for final height per plant in centimeter in both field and pot.

3.13.4. Number of branches per plant

Randomly 10 plants were taken from plot and pots and the number of branches per plant and pots were counted at 30 DAS, 60 DAS and 105 DAS. The average of branch number of three plants was taken for final branch number per plant.

3.13.5. Vigor Index

In pot experiment, 3 plants of each pot were randomly selected, uprooted and root length and shoot length were measured by using centimeter scale after 30 days of sowing. The average number was used. Then vigor index was calculated by using this formula suggested by Abdul- Baki and Anderson (1973):

Vigor Index = (RL+SL) ×GP

Here, RL= Root length (cm)

SL= Shoot length (cm)

GP= Germination percentage

3.13.6. Harvesting of crops

When the plants in the experimental field showed 80% to 90% maturity based on straw color, pod filling, pod color, water content per plant etc. indices. At maturity total plants per plot and pot were harvested and tagged based on plot identity. Total 27 plots were separately harvested for data collection.

3.13.7. Number of pods /plant

After harvesting, plants were collected from different plots to count the number of pods /plants through visual counting

3.13.8. Yield (g/plot/pot)

Yield was counted in both field and pot by weighing total harvested grains per plot. The yield was measured in gram for final data preparation.

3.14. Experimental layout and design

In pot experiment, the experiment was set in a Completely Randomized Design (CRD) with three replications. 12 inches planting pots were used at this experiment. In field experiment, after final land preparation, the field layout was done. The experiment was set in a Randomized Complete Block Design (RCBD) with three replications. The total plot was divided into three blocks each comprising nine (9) plots of 2m X 1.5m size, giving 27 plots. The spaces were kept between blocks was 1m and 0.5m between plots. Planted seeds in five rows per unit plot and the row to row distance was kept 37 cm. The plant to plant distance was maintained as 5 cm and the seeds

were sown in lines in the experimental plots. The seeds were planted at about 4 cm depth in the soil.

Plate 4. Experimental layout of field (left) and pot (right) experiments

3.15. Statistical analysis

The data collected from different parameters were properly compiled and arranged in excel sheets. Appropriate statistical analysis was done by Statistix-10 computer package program. The treatment means were compared with LSD (Least Significant Difference) value at 0.05% alpha value.

3.16. Tagging of plants

In pot experiment, ten plants per pot were maintained for each treatment combinations including control. Randomly three plants were selected from each pot tagged for data collection. Mean values determined to get rating score of each treatment. In field experiment, randomly 10 plants were selected from each plot tagged for data collection.

3.17. Pot experiments

Experiment 1: Seed treatment of lentil for *in vitro* evaluation of commercial bio-fungicides against foot and root rot disease of lentil

Experiment 2: *In vitro* evaluation of soil treatment by commercial bio-fungicides against foot and root rot disease of lentil

Experiment 3: *In vitro* evaluation of commercial bio-fungicides against foot and root rot disease of lentil through seed and soil treatment

3.17.1. Soil Collection

Soil was collected from the experimental fields of Sher-e-Bangla Agricultural University, Dhaka-1207.

3.17.2. Soil solarization

Collected soil was mixed with cow dung properly in ratio 1:3. Then the soil was kept in the open place for solarization by direct sun light for 10 days.

3.17.3. Preparation of pots

Solarized soils were put in the plastic pot of 12″ height and 10″ width (Plate-5). In the bottom of the pot 2 cm hole was made to minimize the losses of excess water. Each pot was filled with 2/3rd of solarized soil and the pots were arranged according to experimental design.

Plate 5. Preparation of pots

3.17.4. Treatments

In experiment 1, seed treatment by commercial bio-fungicides was evaluated against foot and root rot disease of lentil. One chemical check was used to compare the treatments. Altogether 9 treatments including control were assessed in this experiment:

T_0= Control

T_1= G-derma-P @ 5 g/liter water

T_2= Bio- derma @ 5 g/liter water

T_3= Decoprima @ 5 g/liter water

T_4= Recharge @ 5 g/liter water

T_5=Tricost @ 5 g/liter water

T_6= G-derma-L @ 5ml/liter water

T_7=Terrabio @ 5 g/liter water

T_8= Autostin @ 5 g/liter water

In Experiment 2, soil treatment by commercial bio-fungicides was evaluated against foot and root rot disease of lentil. One chemical check was used to compare the treatments. Altogether 11 treatments including control were assessed in this experiment:

T_0= Control

T_1= G-derma-P @ 2 g/liter water

T_2= Bio-derma-P @ 2 g/liter water

T_3= Decoprima @ 2 g/liter water

T_4= Recharge @ 2 g/liter water

T_5= Bio-derma (Peat soil) @100 g/pot

T_6= G-derma-L @ 2 ml/liter water

T_7=Terrabio @ 2 g/liter water

T_8= Formaldehyde @ 5% solution

T_9= Decohumat @ 2 g/liter water

T_{10}= Tricost @ 2 g/liter water

For experiment 3, seed and soil treatment by commercial bio-fungicides was evaluated against foot and root rot disease of lentil. Altogether 8 treatments including control were assessed in this experiment:

T_0= Control

T_1= G-derma-L @ 5ml/liter water (seed treatment) + 2ml/liter water (soil treatment)

T_2= G-derma-P @ 5g/liter water (seed treatment) + 2g/liter water (soil treatment)

T_3= Bio-derma @ 5g/liter water (seed treatment) + 2g/liter water (soil treatment)

T_4= Decoprima @ 5g/liter water (seed treatment) + 2g/liter water (soil treatment)

T_5=Recharge @ 5g/liter water (seed treatment) + 2g/liter water (soil treatment)

T_6= Terrabio @ 5g/liter water (seed treatment) + 2g/liter water (soil treatment)

T_7=Tricost @ 5g/liter water (seed treatment) + 2g/liter water (soil treatment)

3.17.5. Procedure of preparation and application of treatments

a. Seed treatment

In case of G-derma (powder), Decoprima, G-derma (LDS), Recharge, Autostin 5g treatments mixed with 1 liter water for making a solution. Then seeds (20gm) were dipped with the solution properly. The seeds were shaken properly with the suspension and kept the seeds in the shade for an hour for drying before seed sown. For Bio-derma preparation, 1 ml Bio-derma solution was prepared by using 5gm Bio-derma powder. After that seeds were mixed with the solution properly and kept the seeds in the shade for an hour for drying before sown. For Tricost preparation, at first made a solution of 5 g Tricost powder and then mixed with 20 g seeds by that solution. Seed treatment was done before sown and kept the seeds in the shade for an hour for drying. For Terrabio preparation, 5 g Terrabio was mixed with 1 liter water and the mixing jar was remained open for 48 hours. Then that solution was mixed with the seeds and kept the seeds in the shade for an hour for drying before sown.

b. Soil treatments

In case of G-derma (powder), G-derma (LDS), Bio-derma, Recharge, Tricost 2 g was mixed with 1 liter of water. Then 6-8 liter water was mixed with that solution and drenching in the soil of the pots. During this time, water was sprinkled from time to time as required to keep the humidity constant and after 3-4 days the soil was reversed properly. In case of Decoprima, 2 g Decoprima was mixed with 1 liter water and kept it 48 hours and kept the cap of the bottle open. Then 6-8 liter water was mixed with that solution and sprayed in the soil of the pots. Bio-derma (peat soil), 100 g/pot (peat soil) was mixed properly with the soil. For Terrabio preparation, 2 g Terrabio was mixed with 1 liter water and the mixing jar was remained open for 48 hours. Then 6-8 liter water was mixed with that solution and sprayed in the soil of the pots. In case of Formaldehyde, 5% formalin solution was prepared in a container and drenched the soil @ 4-5liter water per square meter soil surface to saturate it up to a depth of 15-20 cm. Drench soil kept cover with double polyethylene sheet for 3 days. The margin of the polyethylene sheet was air tied by the wet soils and bricks. The soil was uncovered and pulverized enough and kept for 7 days to release the gas of formalin. After the smell of the formalin removed completely, the soil was transferred into the pot. For Decohumat preparation, 2 g Decohumat was mixed with 1 liter water and the mixing jar was remained open for 48 hours. Then 6-8 liter water was mixed with that solution and sprayed in the soil of the pots.

c. Seed and Soil treatments

Treatments were prepare and applied as per procedure of 3.17.5. (A and B).

3.17.6. Inoculum Preparation

In pot experiments, *Sclerotium rolfsii* the causal organism of foot and root rot disease of lentil was isolated from the infected lentil plants. In this experiment inoculum was prepared through mass culture. The isolates of *S. rolfsii* was multiplied on barley grains (Gupta and Kolte, 1982). For preparation of mass culture of *Sclerotium rolfsii* barley grains were soaked in water containing 2% sucrose solution for overnight, removed the excess solution. These grains were placed in 500ml conical flask @ 20g and autoclaved at 121.6℃ temperature, under 15 PSI pressure for 15 minutes. The conical flasks were allowed to cool at room temperature and were inoculated with 5mm discs culture of *Sclerotium rolfsii* grown on PDA. Seven discs per flask were added and flasks were placed at room temperature for three months.

Plate 6. Mass culture of *Sclerotium rolfsii* on barley grain

3.17.7. Artificial inoculation

At seedling stage (20 DAS), isolates of causal pathogen (*Sclerotium rolfsii*) were inoculated in the selected plants. The plants were prepared for inoculation by removing surrounding soil of plant with 2cm depth. A tea spoon of inoculum was added to the soil around the plant base and covered with removed soil and then irrigation was done after inoculation to keep moist conditions.

3.18. Field experiment

***In vivo* evaluation of seed treatments by commercial bio-fungicides against foot and root rot disease of lentil in field**

3.18.1. Treatments

In field experiment, seed treatment by commercial bio-fungicides was evaluated against foot and root rot disease of lentil. One chemical check was used to compare the treatments. Altogether 9 treatments including control were assessed in this experiment:

T_0= Control

T_1= G-derma-P @ 5 g/liter water

T_2= Bio-derma @ 5 g/liter water

T_3= Decoprima @ 5 g/liter water

T_4= Recharge @ 5 g/liter water

T_5= Tricost @ 5 g/liter water

T_6= G-derma-L 5 ml/liter water

T_7= Terrabio @ 5 g/liter water

T_9= Autostin @ 5 g/liter water

3.18.2. Diseased plant sample collection

Diseased plant exhibited different types of typical symptoms were collected from pots and field. The samples were carried to the Plant Disease Clinic of Sher-e-Bangla Agricultural University in individual snap locked plastic bags. The collected samples were examined for visual symptoms as well as isolation of causal organisms.

3.18.3. Isolation and identification of causal organism(s)

Isolation and identification of causal organism was done by the following methods:

a. By symptomological study (Visual Assessment)

In this study, the development of symptoms was closely observed to confirm disease. The diseased plants were closely and carefully examined by magnifying glass to observe the disease symptoms development, sign of the pathogen, source of infection, mode of dissemination and favorable environment. Idea about causal organism was taken from those information (Bakr, 1986; Punja *et al*., 1985; Singh, 1982; Ahmed, 1980).

b. By growing on moist blotter paper (Incubation Method)

The diseased roots were cut into pieces and surface sterilized with 70% Ethanol for 30 seconds. Then the sodium hypochlorite (NaOCl) for 30 seconds and washed three times in sterile distilled water each for 1 min. Then the cut pieces were incubated at room temperature for seven days. When the fungus grew well mycelium and sclerotia were observed by visual observation. The identification of the causal organism was done with the

help of relevant literature (Bakr, 1986; Punja *et al*., 1985; Singh, 1982; Ahmed, 1980).

Plate 7. Growing fungi in moist blotting paper

c. By growing on culture media (Tissue Plating Method)

Infected roots were cut into small pieces and some samples were surface sterilized with 70% Ethanol for 30 seconds. Then 1% sodium hypochlorite (NaOCl) for 30 seconds and washed three times in sterile distilled water each for 1 min. Some samples were sterilized with 37.5% chlorox for 30 seconds washed three times in sterile distilled water each for 1 min. The surface sterilized roots were placed on blotter paper and Potato Dextrose Agar (PDA) medium (Mehrota and Aggarwal, 2003) and incubated at 25 ± 2^{0}C for 6-7 days. The plates containing root pieces were incubated at room temperature for three days. When the fungus grew well, the organism

was re-cultured by mycelium or sclerotia to obtain pure culture. The identification of the causal organism was done with the help of relevant literature (Bakr, 1986; Punja *et al*., 1985; Singh, 1982; Ahmed,1980).

Mycelial growth from each developing colony were cultured on PDA to get pure culture. The causal organism was isolated, identified and recorded. The pathogen was identified from all infected samples, (Agrios, 2005).

Plate 8. Culture of *Sclerotium rolfsii* on PDA medium

CHAPTER 4
RESULTS AND DISCUSSION

Among the different disease of lentil, foot and root rot caused by *sclerotium rolfsii* is one of the major and severe soil borne disease responsible for high yield losses. The present study were carried out to evaluate seed treatment, soil treatment and seed and soil treatment together by commercial bio-fungicides during *rabi* season, 2019-2020. The experimental results are presented here.

4.1. Symptoms of foot and root rot disease of lentil

This disease affects mainly in roots, leading to poor emergence of seedlings, stunted growth of plants and reduced yields. Symptoms include sunken lesion and brown or black discoloration on roots, shrinking root system and root decay. If they develop at all, nodules are less numerous, smaller and pale in color. In plants growing from infected seeds, seedlings can blight shortly after emergence. The plants that survive are chlorotic and have poor vigor. Plants infected during late stages of development show stunted growth. Opportunistic pathogens colonize and feed on decaying tissue which makes the symptoms worse. In the field condition the disease often occurs in patches and may expand if conditions are favorable for the pathogens.

Plate 9. Diseased plant showing mycelium of fungus at collar region

4.2. Identification of causal organism

Sclerotium rolfsii was isolated in PDA medium from infected plant sample. After well growth of *Sclerotium rolfsii* on Potato Dextrose Agar (PDA) medium, the organism was re-cultured by mycelium and sclerotia to obtain pure culture. It was observed that, mycelial growth of causal organism was formed from second days after incubation (DAI) and it took a week to fill the whole petri dish with mycelium of *Sclerotium rolfsii*. Mustard seed like sclerotia was formed in the pure culture of causal organism within the two weeks of incubation. Ahmed (1980) found that *Sclerotium rolfsii* is a facultative saprophyte and can survive generation to generation by formation of brown sclerotia. Punja *et al*., (1958) found that the mycelial growth rate of *Sclerotium rolfsii* was maximized under optimum temperature.

2 DAI

4 DAI

6 DAI

8 DAI

Plate 10. Pure culture of *Sclerotium rolfsii* at different days after incubation (DAI)

Pot Experiments in Net House

4.3. Seed treatment of lentil for *in vitro* evaluation of commercial bio-fungicides against foot and root rot disease of lentil

4.3.1. Effect of seed treatments by different *Trichoderma* formulations on disease incidence of foot and root rot disease of lentil at different days after inoculation (DAI)

Disease incidence at different days after inoculation had been recorded on the basis of visible typical symptoms. Nine treatments were compared with each other regarding disease incidence those were recorded at 10, 20, 30 and 60 days after inoculation (DAI).

At 10 DAI, T_5 (Tricost) showed the lowest disease incidence (4.31%) that was statistically similar with T_7 (5.89%), T_6 (6.13%), T_3 (6.27%), T_2 (6.48%) and T_4 (7.27%). The highest disease incidence (47.20%) was recorded in treatment T_0 (control). At 20 DAI, the similar trends were observed where the lowest disease incidence was recorded in T_5 (5.74%) and the highest disease incidence (51.61%) was recorded in control. At 30 DAI, the highest incidence of disease was also recorded from T_0 (53.80%) which incurred the highest yield loss and the lowest disease incidence was recorded in T_5 (Tricost), (6.48%).

At 60 DAI the final disease incidence was recorded where the lowest diseases incidence was recorded in Tricost (6.48%) in T_5 treatment that contributed the highest yield of lentil grain and T_2, T_3, T_4, T_6, T_7 showed

statistically similar results with T_5. Control treatment T_0 incurred the highest yield loss and highest disease incidence (54.9%).

Table 3: Effect of seed treatments by *Trichoderma* formulation on disease incidence (%) in lentil plant at different days after inoculation (DAI) in pot

Treatments	Disease Incidence (%)			
	10 DAI	20 DAI	30 DAI	60 DAI
T_0 (Control)	**47.20 a**	**51.61 a**	**53.80 a**	**54.91 a**
T_1 (G-derma-P)	9.14 b	9.14 b	10.97 bc	11.87 bc
T_2 (Bio-derma)	6.48 bc	6.48 b	7.28 bc	7.28 d
T_3 (Decoprima)	6.27 bc	6.27 b	7.84 bc	9.37 cd
T_4 (Recharge)	7.27 bc	7.27 b	10.05 bc	10.05 b-d
T_5 (Tricost)	**4.31 c**	**5.74 b**	**6.48 c**	**6.48 d**
T_6 (G-derma-L)	6.13 bc	6.89 b	6.89 bc	6.89 d
T_7 (Terrabio)	5.89 bc	6.63 b	7.32 bc	7.32 d
T_8 (Autostin)	9.59 b	9.59 b	11.52 b	13.49 b
CV (%)	22.64	22.85	20.41	15.29
LSD (0.05)	4.41	4.77	4.75	3.72

In a column having same letters do not differ significantly at 5% level of significance by LSD.

4.3.2. Effect of seed treatments by *Trichoderma* formulation on growth parameters in lentil plant at different days after sowing (DAS) in pot

a. Seed germination (%)

Nine treatments were compared with each other for percent seed germination. Among the treatment, the lowest seed germination was counted in control pot (50.66%). Whereas, Tricost treated pots T_5 (77.33%) showed the highest germination percentage followed by T_7 (75.66%), T_6 (72.66%), T_3 (70.66%), T_2 (68.33%), T_1 (61.00%), T_4 (59.66%) and T_8 (57.66%).

b. Plant height (cm)

The effect of seed treatments on the plant height was differed significantly with some extent. Nine treatments were compared with each other for plant height recorded at 30, 60 105 days after sowing (DAS). At 30 DAS the lowest plant height (12.50cm) was recorded in control treatment (T_0) where T_5 (Tricost) showed the highest plant height which was (16.16cm). At 60 DAS the similar trends of results were found where the lowest plant height was recorded in T_0 (14.61cm) and the highest plant height (20.77cm) was recorded in T_5 (Tricost) treatment. At 105 DAS final plant height was recorded and it showed whereas T_5 (Tricost) showed the highest plant height (31.38cm) which was statistically similar with T_2, T_3, T_4, T_6 and T_7. The lowest plant height in (23.22cm) was recorded in control treatment (T_0). Thus, Tricost (T_5) treatment showed best result in case of plant height of lentil plant against foot and root rot disease of lentil.

c. Number of branch/plant

Number of branch was counted by visual observation at 30, 60 and 105 days after sowing (DAS). At 30 DAS treatment T_5 (Tricost) showed the highest branch number (3.66). The second highest number of branches were recorded in case of treatment T_3 (3.33) which was statistically identical to T_4. The third highest branch number were recorded in case of treatment T_0 (3.00cm) which was statistically similar to $T_{1,}$ $T_{2,}$ T_6, T_7 and T_8. Thus, treatment T_5 (Tricost) is the best in increasing of branch number.

At 60 das T_5 treatment showed the highest branch number (5.33) and T_0 (control) showed the lowest branch number which was (4.00). At 105 DAS treatment T_5 and T_2 showed the highest branch number (6.33). The second highest branch number was recorded in treatment T_6 and T_8 (6.00). Treatment T_3 showed third highest number of branch (5.66). Treatments T_0, T_1, T_4 and T_7 treatments were showed similar branch number which was (5.3).

d. Root length (cm)

In case of root length, T_5 (Tricost) showed the highest root length (16.16 cm) followed by T_7 (15.66 cm), T_2 (15.10 cm), T_1 (14.83 cm), T_3 (14.66 cm), T_8 (14.66 cm), T_4 (14.53 cm) and lowest root length showed by T_0 (12.50cm).

e. Shoot length (cm)

In root length of plant, T_5 (Tricost) showed the highest shoot length (16.16 cm) which was statistically similar with T_7 (Terrabio). The lowest shoot length was observed in control treatment T_0 (12.5 cm).

f. Vigor index

Among the treatments, control treatment showed the lowest vigor index which was (908.13) and the highest vigor index was recorded in treatment T_5 (1770.5) that was statistically similar with the treatments of $T_{2,}$ T_3 T_6 T_7. Thus, T_5 (Tricost) showed the best results compared with other treatments.

Table 4. Effect of seed treatments by *Trichoderma* formulation on growth parameters in lentil plant at different days after sowing (DAS) in pot

Treatments	Germination (%)	Plant height (cm)			No. of branch/plant			Root length (cm)	Shoot length (cm)	Vigor index
		30 DAS	60 DAS	105 DAS	30 DAS	60 DAS	105 DAS			
T_0 =Control	**50.66 d**	**12.50** d	**14.61 e**	**23.2 d**	3.0 b	4.00 b	5.3 a	**5.4 b**	**12.5 d**	**908.13 d**
T_1=G-derma-P	61.00 c	14.83 bc	14.83e	29.39 c	3.00 b	4.33 ab	5.3 a	6.76 ab	14.8 bc	1319.6 bc
T_2=Bioderma	68.33 b	15.10 bc	18.50 bc	28.89 bc	3.00 b	5.00 ab	6.3 a	6.66 ab	15.10 bc	1487.6 ab
T_3=Decoprima	70.66 d	14.67 bc	18.44 bc	28.94 bc	3.33 ab	4.66 ab	5.6 a	9.16 a	14.66 bc	1653.3 a
T_4= Recharge	59.66 c	14.53 c	19.33 b	29.11 bc	3.33 ab	4.33 ab	5.3 a	6.66 ab	14.53 c	1266.0 bc
T_5 = Tricost	**77.33 a**	**16.16 a**	**20.77 a**	**31.38 ab**	**3.66 b**	**5.33 a**	**6.3 a**	**5.20 b**	**16.16 a**	**1770.5 a**
T_6=G-derma-L	72.66 ab	15.00 bc	18.00 c	29.88 a-c	3.00 b	4.66 ab	6.0 a	6.53 ab	15.00 bc	1552.3 a
T_7=Terrabio	75.66 a	15.66 ab	19.11 bc	32.11 a	3.00 b	4.33 ab	5.3 a	7.66 ab	15.66 ab	1653.3 a
T_8=Autostin	57.66 c	14.66 bc	16.50 d	29.66 a-c	3.00 b	4.66 ab	6.0 a	4.26 b	14.66 bc	1121.6 cd
CV (%)	4.41	4.07	4.36	8.88	10.59	15.11	12.9	31.05	4.07	12.36
LSD (0.05)	4.87	1.03	1.32	2.54	0.57	1.19	1.19	3.45	1.03	300.81

In a column having same letters do not differ significantly at 5% level of significance by LSD.

4.3.3. Effect of seed treatments by *Trichoderma* formulation on yield and yield contributing characters in lentil plant at different days after sowing (DAS) in pot

Effect of different seed treatments had been compared on the basis of yield and yield contributing characters of lentil viz. number of pods per plant, yield/ plant (g) and yield/pot (g).

a. Number of pod/plant

Number of pod was counted by visual observation after the harvest of the experiment. The highest pod number was counted from T_5 (Tricost) treatment (16.00) and the lowest was conducted T_0 treatment (8.66). The second highest number of pod was recorded from treatments T_4 (14.00). T_7 and T_8 (13.33) treatments were showed third highest number of pod. Treatment T_2 and T_3 showed similar number of pod which was (12.00) and T_1 (10.66) showed rest one. Thus treatment T_5 is the best in increasing total number of pods/ plant.

b. Yield /plant (g)

In case of yield/plant T_5 (Tricost) showed the highest yield/plant (0.50 g). Treatment T_7 and T_8 showed similar result (0.48 g), T_3 and T_6 Showed (0.47 g), T_2 and T_4 (0.45 g) and T_1 (0.43 g). T_0 showed the lowest yield/plant (0.37 g).

c. Yield/pot (g)

The effect of different treatments on yield/pot (g) found to be differed significantly at some extent. The highest yield (12.55g) was recorded in T_5 (Tricost) treatment. The rest of treatment found that T_7 (11.16 g), T_6

(10.88g), T_3 (10.34 g), T_2 (9.2 g), T_4 (9.11 g), T_1 (8.90 g) and treatment T_8 was showed (8.57 g). The lowest yield/pot (6.03g) was recorded in control treatment T_0.

Table 5. Effect of seed treatments by *Trichoderma* formulation on yield and yield contributing characters in lentil plant at different days after sowing (DAS) in pot

Treatments	No. of pods/pot	Yield/plant (g)	Yield/pot (g)
T_0 =Control	**8.66 d**	**0.37 a**	**6.03 d**
T_1=G-derma-P	10.66 cd	0.43 ab	8.90 c
T_2=Bio-derma	12.00 bc	0.45 ab	9.2 c
T_3 =Decoprima	12.66 bc	0.47 a	10.34 b
T_4=Recharge	14.00 ab	0.45 ab	9.11 c
T_5=Tricost	**16.00 a**	**0.5 a**	**12.55 a**
T_6=G-derma-L	13.00 bc	0.47 a	10.88 b
T_7= Terrabio	13.33 b	0.48 a	11.16 b
T_8=Autostin	13.33 b	0.48 a	8.57 c
CV (%)	12.19	13.51	6.46
LSD (0.05)	2.66	0.11	1.06

In a column having same letters do not differ significantly at 5% level of significance by LSD.

In experiment 1, seed treatment with different *Trichoderma* based bio-fungicides showed effectiveness against the disease. Among the treatments, T_5 (Tricost) showed the best performance compared with others treatments against foot and root rot disease of lentil regarding disease incidence, growth parameters and yield contributing characters. Similar results also reported by Jahan *et al*. (2017); Dwivedi and Ganesha (2016). They reported that *Trichoderma* based bio-fungicides gave better performance against foot and root rot disease. Jahan *et al*. (2017) conducted an experiment where from the pot experiment, it was observed that plants treated with bio-pesticide (*Trichoderma* based) reduced Sclerotium rot infection. A field experiment was conducted to investigate the efficacy of *Trichoderma* based bio-pesticide to control Sclerotium rot of sugarbeet. After germination of sugarbeet plants, periodically infections were recorded and results showed that incidence of Sclerotium rot of sugarbeet was very low in bio-pesticide treated plots, where the best performance was recorded with bio-pesticide treatment at 80 kg/ha.

4.4. Experiment 2. *In vitro* evaluation of soil treatment by commercial bio-fungicides against foot and root rot disease of lentil

4.4.1. Effect of soil treatments by *Trichoderma* formulation on disease incidence (%) in lentil plant at different days after inoculation (DAI) in pot

Disease incidence at different days after inoculation during growth period had been recorded on the basis of visible typical symptoms. Eleven treatments were used compared with each other for disease incidence was

recorded at 10, 20, 30 and 60 days after inoculation (DAI). At 10 DAI, the lowest disease incidence was recorded in treatment T_5 Bio-derma (Peat soil) which was (4.17%) proceed by treatments T_4 (4.33%), T_{10} (4.41%), T_3 (4.92%), T_1 (5.95%), T_2 (6.35%), T_6 (6.80%), T_9 (6.98%), T_7 (10.5%) and T_8 (10.86%). The highest disease incidence was recorded in control treatment T_0 (46.77%).

In Table 6, at 20 DAI the highest disease incidence was recorded in T_0 (51.11%) in control treatment and the lowest disease incidence was recorded in treatment T_5 (4.17%). At 30 DAI the highest disease incidence was found in T_0 treatment which was (53.29%) and the lowest was recorded in T_5 (5.17%). Finally, at 60 DAI the lowest disease incidence found in Bio-derma (Peat soil), T_5 (4.84%) which was statistically similar with T_1, T_2 ,T_3 ,T_4, T_6, T_9 and T_{10}.The highest disease incidence was recorded in T_0 (53.29%) in control treatment. Thus, T_5 Bio-derma (Peat soil) was showed the best result among others.

Table 6. Effect of soil treatments by *Trichoderma* formulation on disease incidence (%) in lentil plant at different days after inoculation (DAI) in pot

Treatments	Disease incidence (%)			
	10 DAI	20 DAI	30 DAI	60 DAI
T_0 =Control	**46.77 a**	**51.11 a**	**53.29 a**	**53.29 a**
T_1=G-derma-P	5.95 c	6.95 b-d	6.95 c	6.95 c
T_2=Bio-derma-P	6.35 c	7.26 b-d	7.26 c	9.08 bc
T_3=Decoprima	4.92 c	5.75 cd	6.47 c	6.47 bc
T_4=Recharge	4.33 c	4.43 d	4.33 c	5.12 c
T_5=Bio-derma (peat soil)	**4.17 c**	**4.17 d**	**5.17 c**	**4.84 c**
T_6=G-derma-L	6.80 bc	6.80 b-d	7.78 bc	7.78 bc
T_7=Terrabio	10.5 b	10.55 bc	11.63 b	11.63 b
T_8=Formaldehyde	10.86 b	10.86 b	11.93 b	11.93 b
T_9=Decohumat	6.98 bc	6.98 b-d	6.98 c	6.98 c
T_{10}=Tricost	4.41 c	4.41 d	4.41 c	5.15 c
CV (%)	23.86	26.36	22.25	22.41
LSD (0.05)	4.11	4.83	4.29	4.46

In a column having same letters do not differ significantly at 5% level of significance by LSD.

4.4.2. Effect of soil treatments by *Trichoderma* formulation on growth parameters in lentil plant at different days after sowing (DAS) in pot

a. Seed germination (%)

In case of seed germination among the eleven treatments Bio-derma (Peat soil) showed the highest germination percentage T_5 (77.66%) which was statistically similar with T_4 and T_{10}. Control treatment T_0 (51.00%) showed the lowest germination percentage.

b. Plant height (cm)

The effect of different treatments on the plant height found to be differed significantly with some extent. Eleven treatments were used compared with each other for plant height was recorded at 30 DAS, 60 DAS and 105 DAS. At 30 DAS the lowest plant height (11.38cm) was recorded in treatment T_9 where T_5 showed the highest plant height which was (15.44 cm). At 60 DAS the results were found where the lowest plant height was recorded in T_7 (13.53cm) and the highest plant height (17.50 cm) was recorded in T_5 treatment (Table 7).

At 105 DAS final plant height was recorded and it showed T_5 showed the highest plant height (27.94 cm) which was Bio-derma (Peat soil). The lowest plant height in (21.77cm) was recorded in control treatment (T_0).

c. Number of branch/plant

Number of branch was counted by visual observation at 30 DAS, 60 DAS and 105 DAS. At 30 DAS treatment T_{10} showed the highest branch number (5.66) and the lowest branch number was recorded in treatment T_0 (3.33).The second highest number of branch were recorded in case of treatment T_3 (5.33cm) which was statistically similar to T_4, T_5, T_6, T_7, T_8 and T_9. The third highest branch number were recorded in treatment T_2 (4.66) and T_1 showed (4.00).

At 60 das T_5 treatment showed the highest branch number (7.66) and T_0 (control) shows the lowest branch number which was (4.66). At 105 DAS treatment T_5 and T_4 showed highest branch number (8.00) which was statistically similar. The second highest branch number was recorded in treatment T_7, T_8 and T_9 (7.66). Treatment T_{10} showed third highest number of branch (7.50). Treatments T_0 (control) was showed the lowest branch number which was (5.33).

d. Root length (cm)

In case of root length treatment T_0 showed the lowest root length (4.00 cm). The highest root length showed treatment T_5 which was (8.06 cm) by Bio-derma (Peat soil) followed by T_7 (6.50 cm), T_2 (6.23cm), T_6 (5.63 cm), T_4 (5.40 cm) and T_8 (4.43 cm) respectively.

e. Shoot length (cm)

In shoot length of plant, T_3 showed the highest shoot length (15.28 cm) which was statistically similar with T_5 treatment. Control treatment T_0 showed the lowest shoot length (9.94 cm).

f. Vigor index

In case of vigor index treatment T_0 showed the lowest vigor index which was (736.50) and the highest vigor index was recorded in treatment T_5 (1691.3) by Bio-derma (Peat soil) where T_4 (1493.0) showed statistically similar results. Other treatments were T_3 (1393.0), T_{10} (1296.6), T_2 (1173.5), T_6 (1166.00), T_9 (1035.9), T_1 (1125.0), T_7 (961.00) and T_8 (846.35).

Table 7. Effect of soil treatments by *Trichoderma* formulation on growth parameters in lentil plant at different days after sowing (DAS) in pot

Treatments	Germination (%)	Plant height (cm)			No. of branch/plant			Root length (cm)	Shoot length (cm)	Vigor index
		30 DAS	60 DAS	105 DAS	30 DAS	60 DAS	105 DAS			
T_0= Control	**51.00 e**	**14.00 a-c**	**15.05 bc**	**21.7 d**	**3.3c**	**4.66 d**	**5.33 e**	**4.00 b**	**9.94 f**	**736.5 g**
T_1=G-derma-P	56.00 c-e	12.94 bc	14.04 bc	24.00 b-d	4.00 bc	5.33 cd	6.33 a	5.36 b	12.94b-d	1025.0 d-f
T_2=Bioderma-P	61.33 c	14.55 ab	14.86 bc	24.50 a-d	4.66 ab	5.33 cd	7.00 b-d	6.23 ab	12.89 b-e	1173.5 cd
T_3=Decoprima	68.33 b	13.39 a-c	15.83 b	26.88 ab	5.33 a	6.33 bc	6.66 cd	5.16 b	**15.28 a**	1393.0 b
T_4=Recharge	77.00 a	13.77 a-c	15.66 b	24.63 a-d	5.33 a	7.00 ab	8.00 a	5.40 a	13.39 b	1493.0 ab
T_5=Bioderma-PS	**77.66 a**	**15.44 a**	**17.50 a**	**27.94 a**	5.33 a	6.66 ab	**8.00 a**	**8.06 b**	14.16 ab	**1691.3 a**
T_6=G-derma-L	60.66 cd	14.16 a-c	16.26 ab	25.05 a-d	5.33 a	7.33 ab	7.00 b-d	5.63 b	13.77 bc	1166.0 c-e
T_7=Terrabio	53.00 e	11.93 c	13.53 c	25.72 a-d	5.33 a	**7.66 a**	7.66 ab	6.50 ab	11.60 e	961.0 ef
T_8=Formadehyde	51.66 e	11.94 c	14.83 bc	22.50 cd	5.33 a	6.33 bc	7.66 ab	4.43 b	11.94 de	846.35 fg
T_9=Decohumat	53.33 de	11.38 a-c	15.83 b	23.83 b-d	5.33 a	7.33 ab	7.66 ab	5.33 b	13.38 bc	1035.9d-f
T_{10}=Tricost	75.66 a	12.40 bc	15.20 b	21.88 d	**5.66 a**	7.00 ab	7.50 a-c	4.73 b	12.39 c-e	1296.6 bc
CV (%)	5.90	10.54	6.28	8.81	13.55	11.44	7.77	23.19	5.98	10.57
LDS (0.05)	5.38	2.40	1.63	3.64	1.06	1.25	1.05	2.19	1.30	208.54

In a column having same letters do not differ significantly at 5% level of significance by LSD.

4.4.3. Effect of soil treatments by *Trichoderma* formulation on yield and yield contributing characters in lentil plant at different days after sowing (DAS) in pot

Effect of soil treatments had been compared on the basis of yield and yield contributing characters of lentil viz. no. of pods per plant, yield/ plant (g), yield/pot (g).

a. **Number of pod/plant**

Number of pod was counted by visual observation after the harvest of the plants. The highest pod number was counted from T_5 treatment (14.33) and the lowest was conducted from T_0 (8.66). The second highest number of pod was recorded from treatments T_4 (14.00). T_{10} treatment showed third highest number of pod (13.66). Treatment T_3 showed number of pod which was (13.33). T_2, T_9 showed (13.00), (12.00) respectively. Treatments T_1 and T_7 showed similar number of pod which was (11.66), T_8 showed (11.00).

b. Yield/plant (g)

The effect of different treatments on yield/plant (g) found to be differed significantly at some extent. The lowest yield (0.36 g) was recorded in control treatment T_0 and the highest yield (0.51gm) was recorded in T_5 by Bio-derma (Peat soil) which was statistically similar to treatment T_2, T_4, T_7, T_9 and T_{10}.

c. Yield/pot (g)

The effect of different soil treatments on yield/pot (g) found to be differed significantly at some extent. The highest yield (16.83g) was recorded in T_5 Bio-derma (Peat soil) treatment where treatment T_4 showed statistically

similar result. The lowest yield (7.51 g) was recorded in control treatment T_0.

Table 8. Effect of soil treatments by *Trichoderma* formulation on yield and yield contributing characters in lentil plant at different days after sowing (DAS) in pot

Treatments	No of pod/pot	Yield/plant (g)	Yield/pot (g)
T_0= Control	**8.66 e**	**0.36 e**	**7.51 e**
T_1= G-derma-P	11.66 cd	0.44 cd	12.26 de
T_2= Bio-derma-P	13.00 a-c	0.48 a-c	13.50 c
T_3=Decoprima	13.33 a-c	0.48 a-c	13.83 bc
T_4=Recharge	14.00 a	0.50 ab	16.43 a
T_5=Boi-derma (peat soil)	**14.33 a**	**0.5a ab**	**16.83 a**
T_6=G-derma-L	13.00 a-c	0.48 a-c	13.00 cd
T_7=Terrabio	11.66 cd	0.45 b-d	11.11 ef
T_8=Formaldehyde	11.0 d	0.41 de	10.50 f
T_9=Decohumat	12.00 b-d	0.45 b-d	11.83 de
T_{10}= Tricost	13.66 ab	0.48 a-c	4.76 b
CV (%)	0.10	6.31	5.44
LSD (0.05)	1.91	0.49	1.18

In a column having same letters do not differ significantly at 5% level of significance.

In experiment 2, soil treatment with *Trichoderma* based bio-fungicides showed effectiveness against the disease. Among the treatments T_5, Bio-derma (Peat soil) showed best performance compared with others treatments against foot and root rot disease of lentil regarding disease incidence, growth parameters and yield contributing characters. The present findings are supported by previous research reports Khalequzzaman (2016), Pratibha *et al*. (2016). Pratibha *et al*. (2016) reported that to avoid extensive use of pesticides, biological formulation developed from *Trichoderma harzianum* (Th3) was used as an environment friendly option. In the dual culture method, *T. harzianum* Th3 showed around 62% inhibition against *S. rolfsii*. Seed treatment, soil application and drenching with *T. harzianum* Th3 showed minimum disease incidence of 21.6% and 12.6%. Thus it reduces the disease incidence by 66.3 to 78.1% as compared with control. In the same way, there was significant response of application of *T. harzianum* Th3 was observed on plant growth promoting parameters at field level to increase number of pods per plant and shelling % and thereby produces maximum yield of 3.37 and 3.6 t/ha respectively.

4.5. Experiment 3: *In vitro* evaluation of commercial bio-fungicides against foot and root rot disease of lentil through seed and soil treatment

4.5.1. Effect of seed and soil treatments by *Trichoderma* formulation on disease incidence (%) in lentil plant at different days after inoculation (DAI) in pot

Disease incidence at different days after inoculation during growth period had been recorded on the basis of visible typical symptoms. Eight treatments were compared with each other for disease incidence recorded at 10, 20, 30 and 60 DAI. At 10 DAI the highest disease incidence was recorded in control treatment T_0 (41.71%) and the lowest disease incidence was recorded in treatment T_4 which was (4.20%) followed by treatments T_3 (5.69%), T_6 (6.93%), T_2 (7.15%), T_7 (7.18%), T_5 (7.67%) and T_1 (9.01%). At 20 DAI the highest disease incidence was recorded in T_0 (44.72%) in control and the lowest disease incidence was recorded in treatment T_4 (4.20%). At 30 DAI the highest disease incidence was found in T_0 treatment which was (48.92%) and the lowest recorded in T_4 (7.14%).

Finally at 60 DAI the lowest disease incidence found in Decoprima, T_4 (4.87%) which was statistically similar with T_3, and T_6. The highest disease incidence was recorded from control treatment (48.92 %). Thus, Decoprima is the best among all other treatments against foot and root rot disease of lentil in case of decreasing disease incidence.

Table 9. Effect of seed and soil treatments by *Trichoderma* formulation on disease incidence (%) in lentil plant at different days after inoculation (DAI) in pot

Treatments	Disease incidence (%)			
	10 DAI	20 DAI	30 DAI	60 DAI
T_0 =Control	**41.71 a**	**44.72 a**	**48.92 a**	**48.92 a**
T_1=G-derma- L	9.01 b	9.01 b	11.71 b	11.71 b
T_2=G-derma-P	7.15 bc	7.15 bc	7.94 c	8.78 bc
T_3=Bio-derma	5.69 bc	6.43 bc	7.20 d	7.14 cd
T_4=Decoprima	**4.20 c**	**4.20 c**	**7.14cd**	**4.87 d**
T_5=Recharge	7.67 bc	7.67 bc	8.44 bc	9.97 bc
T_6=Terrabio	6.93 bc	6.93 bc	7.69 cd	8.42 b-d
T_7=Tricost	7.18 bc	7.90 bc	10.06 bc	10.79 b
CV (%)	22.07	20.55	15.75	15.00
LSD (0.05)	4.27	4.18	3.61	3.59 b

Mean followed by the same letters in a column do not differ significantly at 5% level of significance by LSD.

4.5.2. Effect of different seed and soil treatments by *Trichoderma* formulation on different growth parameters in lentil plant at different days after sowing (DAS) in pot

a. Germination (%)

In case of germination percentage T_0 (54.33%) showed the lowest germination percentage from control treatment. Decoprima, T_4 (79.33%) showed the highest germination percentage where T_7 showed statistically similar results. Others treatments were T_3 (77.66%), T_7 (77.33%), T_5 (72.33%), T_6 (72.00%), T_2 (70.00%) and T_1 (61.66%) respectively.

b. Plant height (cm)

The effect of different treatments on the plant height found to be differed significantly with some extent. Plant height was recorded at 30 DAS, 60DAS and 105 DAS. At 30 the lowest plant height (14.00cm) was recorded in treatment T_0 (control) where T_4 (Decoprima) showed the highest plant height which was (23.38 cm). At 60 DAS the results was found where the lowest plant height was recorded in T_0 which refers control (16.49cm) and the highest plant height (26.00cm) was recorded in T_4 treatment which was statistically similar to T_3.

At 105 DAS final plant height was recorded in T_4 (Decoprima) showed the highest plant height (41.05 cm) which was statistically similar with T_3 treatment The lowest plant height in (28.77cm) was recorded in treatment (T_0). Thus, Decoprima is the best among the treatments in increasing plant height.

c. Number of branch/plant

Number of branch was counted by visual observation at 30 DAS, 60 DAS and 105 DAS. At 30 DAS treatment T_4 showed the highest branch number (3.36cm). The second highest number of branch were recorded in case of treatment T_3 (3.33cm). Then the highest branch number were recorded in treatment T_5 (2.66cm), T_7 (2.65 cm), T_6 (2.55 cm), T_0 and T_1 (2.44 cm) and T_2 showed (2.33cm). At 60 DAS Decoprima ($T_{4)}$ treatment showed the highest branch number (5.33cm) and T_0 (control), T_2 and T_6 showed the lowest branch number which was (3.77 cm).

At 105 DAS treatment T_4 (Decoprima) showed the highest branch number (6.89cm) where T_1, T_2, T_5 showed statistically similar results with T_4 treatment. The second highest branch number was recorded in treatment T_6 (6.33cm). Control treatment T_0 showed the lowest branch number which was (4.55cm).

d. Root length (cm)

In case of root length treatment T_3 showed the highest root length (8.56 cm) which was followed by T_4 (7.63 cm), T_7 (5.60 cm), T_6 (5.06 cm), T_0 (4.50 cm), T_1 (4.23 cm), T_5 (4.06 cm). The lowest root length showed by treatment T_2 (3.90 cm).

e. Shoot length (cm)

The highest shoot length was observed in treatment Decoprima, T_4 (23.38 cm) which was statistically similar to T_2 (22.11 cm). The lowest shoot length was showed by treatment control treatment T_0 (14.0 cm).

f. Vigor index

In case of vigor index treatment T_0 showed the lowest vigor index which was (1004.4) and the highest vigor index was recorded in treatment Decoprima, T_4 (2462.9) where T_3 (2449.1) showed statistically similar results. Others treatments were T_7 (1777.2), T_2 (1681.7), T_6 (1677.2), T_5 (1566.8) and T_1 (1507.0) respectively.

Table 10. Effect of different seed and soil treatments by *Trichoderma* formulation on growth parameters in lentil plant at different days after sowing (DAS) in pot

Treatments	Germination (%)	Plant height (cm)			No. of branch/plant			Root length (cm)	Shoot length (cm)	Vigor index
		30 DAS	60 DAS	105 DAS	30 DAS	60 DAS	105 DAS			
T_0=Control	**54.33 d**	**14.0 a**	**16.49 e**	**28.7 d**	2.44 c	3.78 c	**4.55 c**	4.50 bc	**14.00 e**	**1004.4 d**
T_1=G-derma-L	61.66 c	20.22 b	23.00 bc	34.27 b	2.44 c	4.00 c	4.89 bc	4.23 bc	20.23 b	1507.0 c
T_2=G-derma-P	70.00 b	20.11bc	23.57 b	33.33 b	**2.33 c**	3.78 c	4.89 bc	**3.90 c**	22.11 bc	1681.7 bc
T_3=Bio-derma	77.66 1	22.91 a	25.93 a	39.50 a	3.33 ab	4.66 ab	5.89 a	**8.56 a**	22.04 a	2449.1 a
T_4=Decoprima	**79.33 a**	**23.38 a**	**26.00 a**	**41.05 a**	**3.36 a**	**5.33 a**	**6.89 bc**	7.63 a	**23.38 a**	**2462.9 a**
T_5=Recharge	72.33 b	17.44 d	20.66 d	32.38 bc	2.66 bc	**3.77 c**	4.77 bc	4.06 c	17.44 d	1566.8 bc
T_6= Terrabio	72.00 b	18.22cd	21.50 cd	33.50 b	2.55 c	3.78 c	6.33 a	5.06 bc	18.21 cd	1677.2 bc
T_7=Tricost	77.33 a	17.33 d	22.0 b-d	30.72 cd	2.65 bc	3.89 bc	5.11 b	5.60 b	17.33 d	1773.8 b
CV (%)	3.81	5.77	4.67	4.05	15.32	12.35	5.24	31.05	4.07	12.36
LDS (0.05)	4.66	1.91	1.81	2.39	0.73	0.88	0.46	1.41	1.90	207.71

Mean followed by the same letters do not differ significantly at 5% level of significance by LSD.

4.5.3. Effect of seed and soil treatments by *Trichoderma* formulation on yield and yield contributing characters in lentil plant at different days after sowing (DAS) in pot

Effect of different seed and soil treatment had been compared on the basis of yield and yield contributing characters of lentil viz. number of pods per plant, yield/ plant (g), yield/pot (g).

a. Number of pods/plant

Number of pod was counted by visual observation after the harvest of the experiment. The highest pod number was counted from T_4 (Decoprima) treatment (16.00) and the lowest was conducted T_0 treatment (9.66). The second highest number of pod was recorded from treatmentsT_3 (15.33) which was statistically similar with treatment T_4. T_2 treatment showed third highest number of pod (12.33). Treatment T_7, T_1 showed similar number of pod which was (12.00), T_5, T_6 showed (11.66) and (11.33) respectively. Thus, treatment T_4 (Decoprima) is the best in increasing total number of pods/ plant.

b. Yield/plant (g)

The effect of different treatments on yield/plant (g) found to be differed significantly at some extent. The highest yield (0.56 g) was recorded in treatment T_4 (Decoprima). The rest of treatments were found that T_3 (0.54 g), T_7 (0,46 g), T_6, T_2, T_6, showed similar yield/plant (0.45gm) and T_5 showed (0.43 g) respectively. The lowest yield/plant showed in control treatment T_0 (0.39 g).

c. Yield/pot (g)

The effect of different treatments on yield (g) found to be differed significantly at some extent. The highest yield (17.83g) was recorded in T_4 (Decoprima) treatment. The rest of treatments were found that T_6, T_3, T_7, T_5, T_2 and T_1 showed (14.16g), (13.83g),(13.45 g), (13.33 g), (12.76 g) and (9.62g) respectively. The lowest yield (7.53 g) was recorded in control treatment T_0 .Thus, Decoprima (T_4) is the best among all other treatments in yield/pot.

Table 11. Effect of seed and soil treatments by *Trichoderma* formulation on yield and yield contributing characters in lentil plant at different days after sowing (DAS) in pot

Treatments	No of pod/pot	Yield/plant (g)	Yield/pot (g)
T_0=Control	**9.66 c**	**0.39 c**	**7.53 e**
T_1=G-derma- L	12.00 b	0.45 b	9.62 d
T_2=G-dema- P	12.33 b	0.45 b	12.76 c
T_3=Bio-derma	15.33 a	0.54 a	13.83 b
T_4=Decoprima	**16.00 a**	**0.56 a**	**17.83 a**
T_5=Recharge	11.33 bc	0.43 bc	13.33 c
T_6=Terrabio	11.66 b	0.45 b	12.16 bc
T_7=Tricost	12.00 b	0.46 b	13.45 c
CV (%)	9.21	6.72	7.54
LSD (0.05)	1.99	0.05	1.70

In a column having same letters do not differ significantly at 5% level of significance by LSD.

In experiment 3, seed and soil treatment with *Trichoderma* based bio-fungicides showed effectiveness against the disease. Among the treatments T_4 (Decoprima) showed best performance compared with others treatments against foot and root rot disease of lentil regarding disease incidence, growth parameters and yield contributing characters which active ingredients were *Trichodermma* sp., *Geobacillus* sp. and *Streptomyces* sp. also. Thus, *Tricoderma* based bio-fungicides is the best in increasing yield and yield contributing characters, growth parameters and decreasing disease incidence against foot and root rot disease. The results are similar with the finding of Hoque *et al*. (2015), Shahiduzzaman (2015). Hoque *et al*. (2015) stated that, an investigation was carried out to evaluate the six selected isolates of three bio-control agents against foot and root rot pathogens. The pathogens, *Fusarium oxysporum* and *Sclerotium rolfsii* were isolated from foot and root rot infected lentil seedlings. Four isolates of *Rhizobium leguminosarum*, one isolate of *Pseudomonas fluorescens* and one isolate of *Trichoderma harzianum* were used as bio-control agents. In case of *S. rolfsii*, 80% and 37.85% inhibition zone were measured against *P. fluorescens* and *T. harzianum*.

Field Experiment

4.6. Experiment 1: *In vivo* evaluation of seed treatments by commercial bio-fungicides against foot and root rot disease of lentil in field

4.6.1. Effect of seed treatments by *Trichoderma* formulation on disease incidence in lentil plant at different days after inoculation (DAI) in field

Disease incidence at different days after sowing during growth period had been recorded on the basis of visible typical symptoms. Nine treatments were compared with each other for disease incidence recorded at 10, 20, 30 and 60 DAI. At 10 DAI the highest disease incidence was recorded in control treatment T_0 (43.33%) and the lowest disease incidence was recorded in treatment T_5 (Tricost) which was (5.54%) followed by treatments T_7 (8.89%), T_6 (14.44%), T_3 (15.55%), T_2 (18.89%), T_1 (22.22%) and T_4, T_8 showed similar disease incidence (25.55%) shown table 12. At 20 DAI the highest disease incidence was recorded in T_0 (46.66%) in control and the lowest disease incidence was recorded in treatment Tricost, T_5 (6.67%). At 30 DAI the highest disease incidence was found in T_0 treatment which was (47.78%) and the lowest recorded in T_5 (7.78%).

Finally at 60 DAI, the lowest disease incidence found in Tricost, T_5 (10.00%) which was statistically similar with T_7. The highest disease incidence was recorded in T_0 (48.89%) in control treatment.

Table 12. Effect of seed treatments by *Trichoderma* formulation on disease incidence (%) in lentil plant at different days after sowing (DAS) in field

Treatments	Disease incidence (%)			
	10 DAS	20 DAS	30 DAS	60 DAS
T_0=Control	**43.33 a**	**46.66 a**	**47.78 a**	**48.89 a**
T_1=G-derma-P	22.22 b	26.67 b	26.67 bc	27.78 bc
T_2=Bio-derma	18.89 cd	22.22 c	23.33 cd	24.44 cd
T_3=Decoprima	15.55 de	18.69 d	20.00 de	22.22 de
T_4=Recharge	25.55 b	27.78 b	28.89 b	28.89 bc
T_5=Tricost	**5.54 g**	**6.67 f**	**7.78 g**	**10.00 f**
T_6=G-derma-L	14.44 e	17.78 d	17.78 e	18.89 e
T_7=Terrabio	8.89 f	11.04 e	12.15 f	13.33 f
T_8=Autostin	25.55 b	27.78 b	28.89 b	31.11 b
CV (%)	9.63	7.20	8.43	10.36
LSD Value	3.33	2.84	3.45	4.49

In a column having same letters do not differ significantly at 5% level of significance by LSD.

4.6.2. Effect of seed treatments by *Trichoderma* formulation on different growth parameters in lentil plant at different days after sowing (DAS) in field

a. Plant height (cm)

The effect of seed treatments on the plant height found to be differed significantly with some extent. Nine treatments were compared with each other for plant heigh at 30, 60 and 105 DAS. At 30 the lowest plant height (13.94cm) was recorded in treatment T_0 (control) where T_5 (Tricost) showed the highest plant height which was (15.22 cm) that was statistically similar with T_1, T_2, T_4, T_6, T_7 and T_8. At 60 DAS the results were found where the lowest plant height was recorded in T_8 (28.83cm) and the highest plant height (30.88cm) was recorded in T_5 (Tricost) treatment.

At 105 DAS the lowest plant height in (31.60 cm) was recorded in treatment (T_0) whereas T_5 showed the highest plant height (39.80 cm) which was statistically similar with T_1, T_2, T_3, T_4, T_6, T_7 and T_8. Thus, Tricost (T_5) is the best among all other treatments in increasing plant height.

b. Number of branch/plant

Number of branch was counted by visual observation at 30, 60 and 105 DAS. At 30 DAS treatment T_5 (Tricost) showed the highest branch number (2.89). The lowest branch number was recorded in treatment T_3 (2.33).

At 105 DAS treatment T_5 (Tricost) showed the highest branch number (7.22) which was statistically similar with T_1, T_2, T_3, T_4, T_6, T_7 and T_8. Treatment T_0 showed the lowest branch number which was (5.55).

Table 13. Effect of seed treatments by *Trichoderma* formulation on growth parameters in lentil plant at different days after sowing (DAS) in field

Treatments	Plant height (cm)			No. of branch/plant		
	30 DAS	60 DAS	105 DAS	30 DAS	60 DAS	105 DAS
T_0 = Control	**13.94 d**	29.22 bc	**31.60 b**	2.74 ab	4.66 cd	**5.55 b**
T_1=G-derma-P	15.61 a	30.44 ab	39.50 a	2.44 ab	4.78 c	6.78 ab
T_2=Bio-derma	14.93 a-c	28.88 a-c	39.66 a	2.44 ab	4.89 c	6.89 ab
T_3=Decoprina	14.00 cd	28.88 a-c	37.50 a	**2.33 e**	**4.22 d**	6.00 ab
T_4=Recharge	14.72 a-d	29.61 a-c	40.07 a	2.55 ab	5.44 ab	7.00 a
T_5=Tricost	**15.22 ab**	**30.88 a**	**39.80 a**	**2.89 a**	**5.78 a**	**7.22 a**
T_6=G-derma-L	14.71 a-d	29.66 a-c	38.55 a	2.44 ab	5.00 bc	7.22 a
T_7=Terrabio	14.93 a-c	30.50 ab	38.21 a	2.55 ab	5.44 ab	6.44 ab
T_8=Autostin	14.50 b-d	**28.83 c**	38.05 a	2.44 ab	4.55 cd	6.33 ab
CV (%)	3.72	2.80	6.99	11.22	5.95	11.78
LSD (0.05)	0.94	1.44	4.61	0.49	0.51	1.34

In a column having same letters do not differ significantly at 5% level of significance

4.6.3. Effect of seed treatments by *Trichoderma* formulation on yield and yield contributing characters in lentil plant at different days after sowing (DAS) in field

a. Number of pod/plant

Number of pod was counted by visual observation after the harvest of the experiment. The highest pod number was counted from T_5 (Tricost) treatment (43.66) which was statistically similar with T_7. The lowest was counted T_0 treatment (21.66). T_6 treatment showed third highest number of pod (35.66). Treatment T_3 showed number of pod which was (33.00), T_2 (32.66), T_1 (30.00) and T_4 (25.33). Thus, treatment T_5 (Tricost) is the best in increasing total number of pods/ plot.

b. Yield/plot (g)

The effect of different seed treatments on yield/plot (g) found to be differed significantly at some extent. The lowest yield (235.00 g) was recorded in control treatment T_0 and the highest yield (437.67g) was recorded in T_5 (Tricost) treatment. The rest of treatments were found that T_7, T_6, T_3, T_2, T_1, T_4 and T_8 showed (425.33g), (404.00g), (389.67 g), (13.33 g), (363.33 g), (329.00g) and (317.67 g) respectively.

c. Yield/m^2 (g)

The effect of different treatments on yield/m^2 (g) found to be differed significantly at some extent. The highest yield (145.89 g) was recorded in T_5 treatment and the lowest yield (78.3 g) was recorded in control (T_0) treatment. Thus, T_5 is the best in increasing yield/m^2 among all other treatments.

Table 14. Effect of seed treatments by *Trichoderma* formulation on yield and yield contributing characters in lentil plant at different days after sowing (DAS) in field

Treatments	Number of pod/plot	Yield/plot (g)	Yield/m^2 (g)
T_0= Control	**21.66 f**	**235.00 h**	**78.3 h**
T_1=G-derma	30.oo de	339.00 f	113.00 f
T_2=Bio-derma	32.66 cd	363.33 e	121.11 e
T_3=Decoprima	33.00 cd	389.67 d	129.89 d
T_4=Recharge	25.33 ef	329.00 fg	109.67 fg
T_5=Tricost	**43.66 a**	**437.67 a**	**145.89 a**
T_6=G-derma LDS	35.66 bc	404.00 c	134.67 c
T_7=Terrabio	38.66 ab	425.33 b	141.78 b
T_8=Autostin	23.33 f	317.67 g	105.89 g
CV (%)	9.53	1.87	1.88
LSD (0.05)	5.20	11.68	3.89

In a column having same letters do not differ significantly at 5% level of significance by LSD.

In field experiment, *Trichoderma* based bio-fungicides treatment T_5 (Tricost) showed best performance among all others treatment against foot and root rot disease of lentil on disease incidence, growth parameters and yield contributing characters. The literature in favour of seed treatment with *Trichoderma* against soil borne pathogens are available in the previous scientific reports viz., Sultana *et al*. (2015), Kashem *et al*. (2014). Kashem *et*

al. (2014) reported that, ten bio-agents including control were tested in these experiments. Macerated extract of *Fusarium solani* + *Trichoderma harzianum* showed the best result in controlling root rot of lentil with the highest seed germination (100%), radical length (1.56 cm), seedling emergence (95.73%), root length (8.16 cm), shoot length (17.75 cm), number of branches/5 plants (15.56).

4.7. Regression Analysis

4.7.1. Regression coefficient between percent disease incidence and yield of lentil (g/pot) for seed treatment by commercial bio-fungicides in pot experiment

The linear regression analysis found negative relationship between yield with percent diseased incidence. However, yield response to the intensity of the percent disease incidence which can be determined by the regression equation $Y = -0.0965x + 11.007$ (R^2=0.6364). The fitted line plot showed the regression results graphically with equation between the dependent variable of yield and independent variable of percent disease incidence. The equation indicates that yield decreases at the rate of –0.0965 (gram) with an increases of one unit of percent diseased incidence. The R^2 value of 0.6364 indicates that yield can be explained as 63% by the respective function

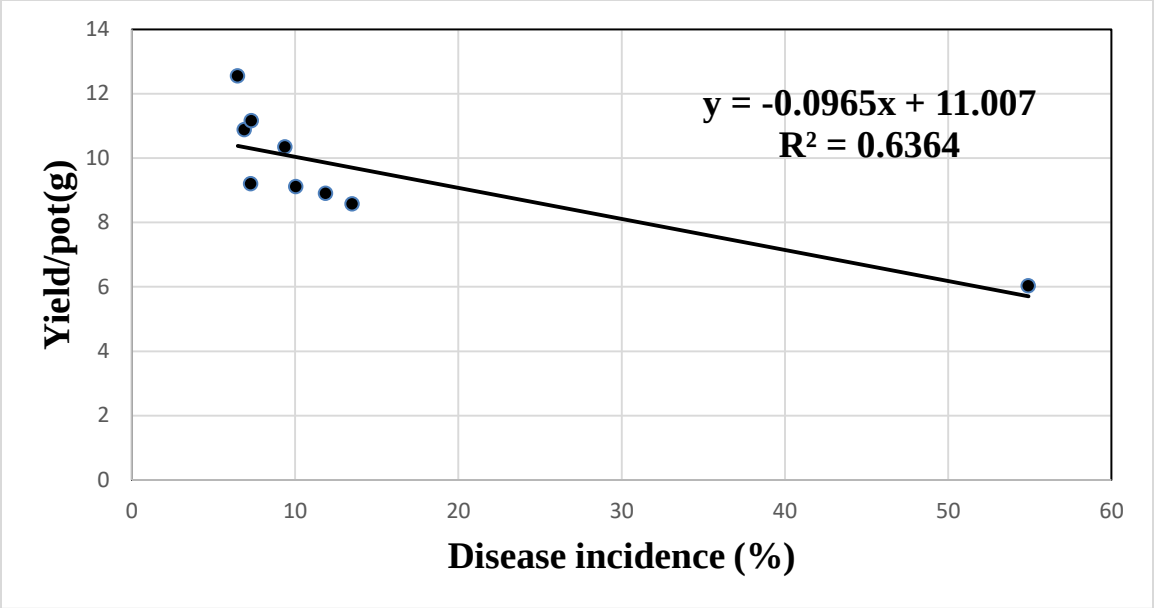


Figure 1: Regression coefficient between percent disease incidence and total yield /pot (g) of lentil for seed treatment in pot

4.7.2. Regression coefficient between percent disease incidence and yield of lentil (g/pot) for soil treatment by commercial bio-fungicides in pot experiment

The linear regression analysis found negative relationship between yield with percent diseased incidence. However, yield response to the intensity of the percent disease incidence which can be determined by the regression equation Y = -0.1459x + 14.584 (R^2=0.5778). The fitted line plot showed the regression results graphically with equation between the dependent variable of yield and independent variable of percent disease incidence. The equation indicates that yield decreases at the rate of –0.1459 (gram) with an increases of one unit of percent diseased incidence. The R^2 value of 0.5778 indicates that yield can be explained as 57% by the respective function.

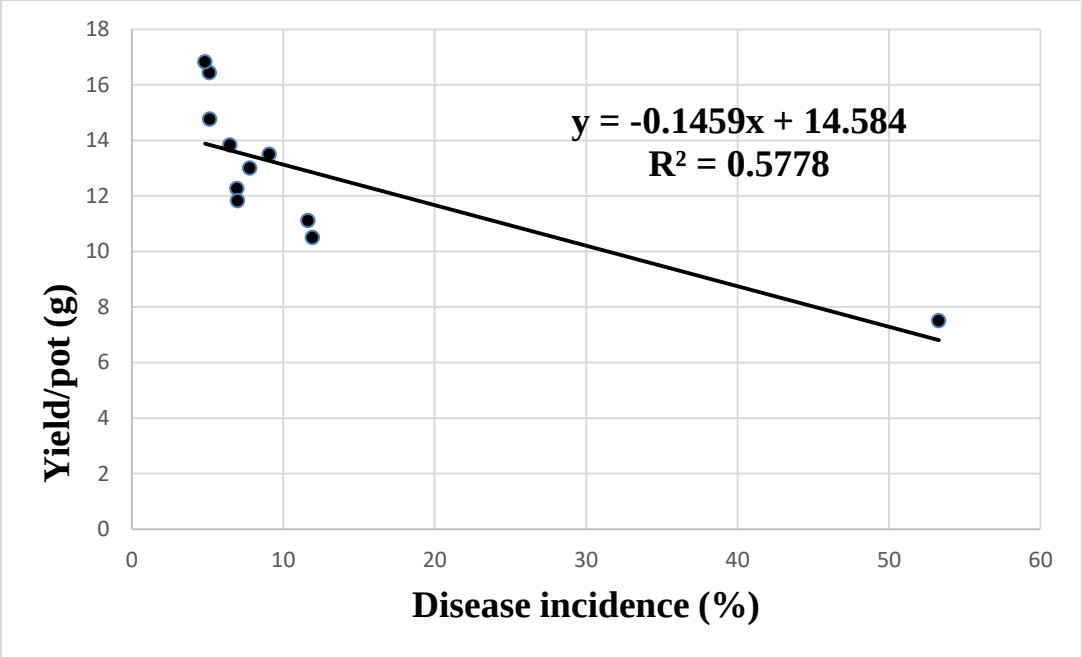


Figure 2. Regression coefficient between percent disease incidence and total yield /pot (g) of lentil for soil treatment in pot

4.7.3. Regression coefficient between percent disease incidence and yield of lentil (g/pot) for seed and soil treatment by commercial bio-fungicides in pot experiment

The linear regression analysis found negative relationship between yield/pot with percent diseased incidence. However, yield response to the intensity of the percent disease incidence which can be determined by the regression equation Y = -0.1676x + 5.1317 (R^2=0.6055). The fitted line plot showed the regression results graphically with equation between the dependent variable of yield and independent variable of percent disease incidence. The equation indicates that yield decreases at the rate of –0.1676 (gram) with an increases of one unit of percent diseased incidence. The R^2 value of 0.6055 indicates that yield can be explained as 60% by the respective function.

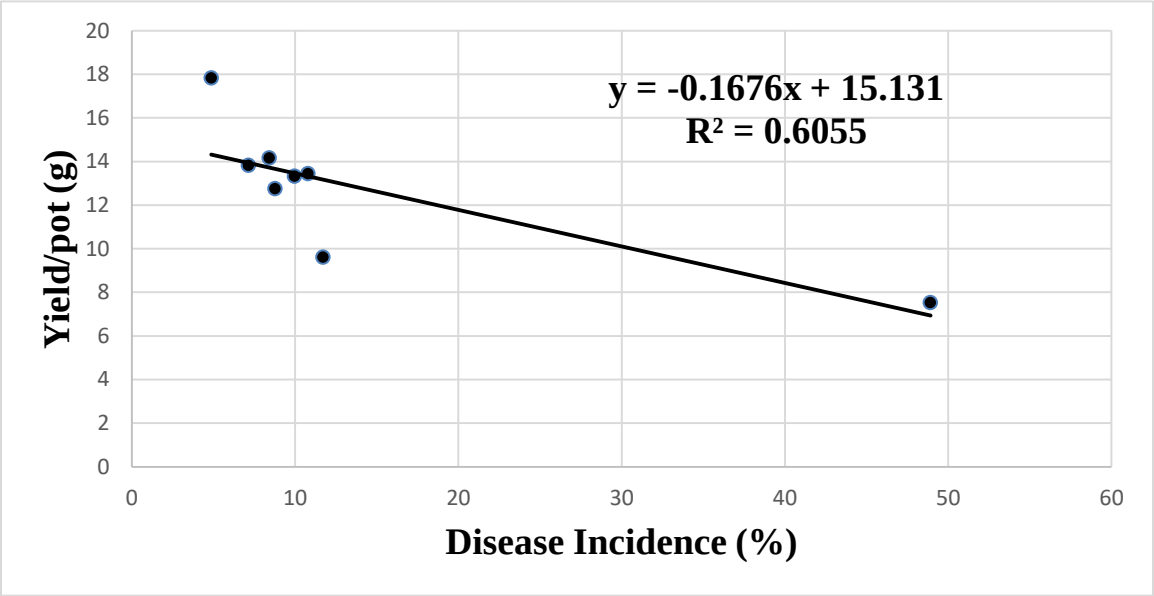


Figure 3. Regression coefficient between percent disease incidence and total yield /pot (g) of lentil for seed and soil treatment in pot

4.7.4. Regression coefficient between percent disease incidence and yield/m^2 of lentil (g/plot) for seed treatment by commercial bio-fungicides in field experiment

The linear regression analysis found negative relationship between yield/m^2 (g) with percent diseased incidence. However, yield/m^2 response to the intensity of the percent disease incidence which can be determined by the regression equation Y = -1.833x + 165.96 (R^2=0.9819). The fitted line plot showed the regression results graphically with equation between the dependent variable of yield and independent variable of percent disease incidence. The equation indicates that yield decreases at the rate of –1.833 (g) with an increases of one unit of percent diseased incidence. The R^2 value

of 0.9819 indicates that yield can be explained as 98% by the respective function.

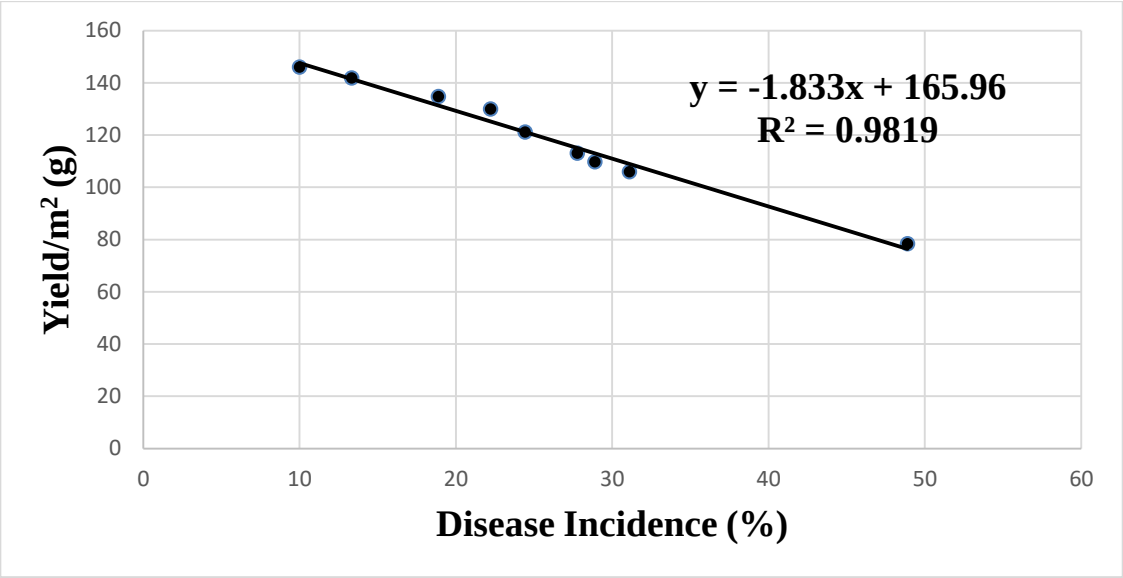


Figure 4. Regression coefficient between percent disease incidence and total yield /m^2 (g) of lentil for seed treatment in field

CHAPTER 5

SUMMARY AND CONCLUSION

Pulses are important legume crops in Bangladesh because of their importance in food, feed, and cropping systems. It contains about twice as much protein as cereals. Pulses have played an important role in sustaining the productivity of soils in Bangladesh for centuries. In Bangladesh, among the diseases of lentil, foot and root rot disease caused by *Sclerotium rolfsii* is a potential threat to lentil production. Considering above facts, this research work was conducted to evaluate seed treatment, soil treatment and seed and soil treatment together by commercial bio-fungicides against foot and root rot disease of lentil.

The experiments were conducted at the Net House and Central Farm of Sher-e-Bangla Agricultural University, Dhaka-1207, during the month of November 2019 to April 2020. Lentil variety BARI Masur -1 (Utfala) was used in this experiment. The experiment was conducted by following CRD and RCBD for pot and field experiments, respectively.

In experiment 1, seed treatment with seven *Trichoderma* based commercial bio-fungicides with one chemical were evaluated viz. G-derma-P, Bio-derma, Decoprima, Recharge, Tricost, G-derma-L, Terrabio and one chemical Autostin with control treatment were investigated and observed their effect on the percent disease incidence of foot and root rot, growth characters and yield contributing characters of lentil and yield of the crop. The effects of most of the nine different treatments were significant in controlling disease of foot and root rot of lentil. Seed treatment with Tricost

(Powder) was found to be the best in controlling disease incidence of foot and root rot of lentil where other treatments showed lower performance.

At 10, 20, 30 and 60 DAI the lowest disease incidence were recorded. At 60 DAI the lowest diseases incidence was recorded in Tricost (6.48%) in T_5 treatment that contributed the highest yield of lentil grain. Control treatment T_0 incurred the highest yield loss and highest disease incidence (54.9%).

The highest number of pods/ plant (16.00) was recorded from T_5 (Tricost) treatment and the lowest number of pods/ plant (8.66) was recorded from T_0 treatment. The highest yield /pot (12.55 g) was recorded from T_5 and the lowest yield/pot (6.03 g) was recorded from T_0 treatment.

The findings of the present study have clearly pointed out that among the fungicides used, Tricost , appeared to be the best in controlling the disease of foot and root rot as well as yield of lentil plant. So the lentil grower can be recommended to use Tricost at 5g/liter water as seed treatment against foot and root rot of lentil caused by *Sclerotium rolfsii*. However, further investigations may be carried out to clarify and justify the results.

In experiment 2, seed treatment with nine commercial bio-fungicides with one chemical were evaluated viz. G-derma (Powder), Bio-derma Powder), Decoprima, Recharge, Bio-derma (Peat soil), G-derma-L, Terrabio, Decohumat, Tricost and one chemical Formaldehyde with control treatment were investigated and observed their effect on percent disease incidence of foot and root rot, growth characters and yield contributing characters of lentil and yield of the crop. The effects of most of the eleven different treatments were significant in controlling disease of foot and root rot of lentil. Soil treatment with Bio-derma (peat soil) @ 100 gm/pot was found to

be the best in controlling disease incidence of foot and root rot of lentil where other treatments showed lower performance.

At 60 DAI the lowest disease incidence (4.84%) was recorded from T_5 whereas the highest disease incidence (53.29%) was observed from T_0 in control treatment.

The highest number of pods/ plant (14.33) was recorded from T_5 treatment and the lowest number of pods/ plant (8.66) was recorded from T_0 treatment. The highest yield /pot (16.83 g) was recorded from T_0 and the lowest yield/pot (7.51 g) was recorded from T_0 treatment. The highest yield/ plant (0.51 g) was recorded from T_5 where the lowest yield/ plant (0.36 g) was recorded from control (T_0).

The findings of the present study have clearly pointed out that among the fungicides Bio-derma (peat soil) appeared to be the best in controlling the disease of foot and root rot as well as yield of lentil plant. So the lentil grower can be recommended to use Bio-derma (peat soil) at 100 g/pot peat soil was mixed properly with the soil as soil treatment against foot and root rot of lentil caused by *Sclerotium rolfsii*. However, further investigations may be carried out to clarify and justify the results.

In experiment 3, six treatments viz. G-derma-L, G-drema-(Powder), Bio-derma, Decoprima, Recgarge,Terrabio, and Tricost with control treatment were investigated and observed on the percent disease incidence of foot and root rot, growth characters and yield contributing characters of lentil and yield of the crop. The effects of most of the nine different treatments were significant in controlling disease of foot and root rot of lentil. Seed and soil treatment with Decoprima was found to be the best in controlling disease

incidence of foot and root rot of lentil where other treatments showed lower performance.

At 60 DAI the lowest disease incidence (4.87%) was recorded from T_4 whereas the highest disease incidence (48.92%) was observed from T_0 treatment.

The findings of the present study have clearly pointed out that among the fungicides Decoprima @ 5g/liter water (seed treatment) + 2g/liter water (soil treatment) appeared to be the best in controlling the disease of foot and root rot as well as yield of lentil plant. So the lentil grower can be recommended to use Decoprima @ 5g/liter water (seed treatment) + 2g/liter water (soil treatment) is suggested as seed and soil treatment against foot and root rot of lentil caused by *Sclerotium rolfsii*. However, further investigations may be carried out to clarify and justify the results.

In field experiment, RCBD design with nine treatments was conducted to evaluation of seed treatment by commercial bio-fungicides against foot and root rot of lentil in field condition. Investigation was carried out to find out the effect of these treatments on the percent disease incidence of foot and root rot, growth characters and yield contributing characters of lentil and yield of the lentil.

The effects of most of the nine different treatments were significant in controlling disease of foot and root rot of lentil. Seed treatment with (Tricost) was found to be the best in controlling disease incidence of foot and root rot of lentil where other treatments showed lower performance.

At 60 DAI the lowest disease incidence (10.00%) was recorded from T_5 whereas the highest disease incidence (48.89%) was observed from T_0 treatment.

The highest number of pods/ plant (43.66) was recorded from T_5 treatment and the lowest number of pods/ plant (21.66) was recorded from T_0 treatment. The highest yield /plot (437.67 g) was recorded from T_5 and the lowest yield/plot (235.00 g) was recorded from T_0 treatment. The highest yield/ m^2 (145.89 g) was recorded from T_5 (Tricost) where the lowest yield/ m^2 (78.3 g) was recorded from control (T_0).

The findings of the present study have clearly pointed out that among the fungicides used Tricost appeared to be the best in controlling the disease of foot and root rot as well as yield of lentil plant both on pot and field condition. So the lentil grower can be recommended to use Tricost at 5g/liter water as seed treatment against foot and root rot of lentil caused by *Sclerotium rolfsii*. However, further investigations may be carried out to clarify and justify the results.

Considering the findings of the present study, further investigation in the following areas may be suggested:

- This experiment may be conducted in different agro-ecological zones of Bangladesh for regional trial before final recommendation
- Further experiment is suggested to evaluate seed and soil treatments together by commercial bio-fungicides in field condition.

CHAPTER 6

REFERENCES

Abdul-Baki, A.A. and Anderson, J.D. (1973). Vigor determination in soybean seed by multiple criteria. *Crop Science*. **13**(6): 630.

Afzal, M.A., Bakr, M.A. and Rahman, M.L. (1999). Lentil cultivation in Bangladesh. Lentil, Blackgram and Mungbean Development Pilot Project, Pulses Research Station, Bangladesh Agricultural Research Institutes, (BARI), Gazipur-1701, Bangladesh.

Agarwal, S.C., Khare, M.V. and Jain, A.C. (1979). Diseases of lentil and their control. Technical Bulletin. Jawaharlal Nehru Krishi Viswa Vidyalaya, Jabalpur, Madhya Pradesh, India. p. 29.

Agrios, G. (2005). Plant Pathology. 5th Edition, Elsevier Academic Press, Amsterdam, The Netherlands.

Ahmed, F. (1980). Control of foot and root rot disease of wheat. M.S. Thesis, Department of Plant Pathology, Bangladesh Agricultural University, Mymensingh, Bangladesh.

Ahmed, H.U., Shahjahan A.K.M. and M.M. Rahman, (1985). A comparison of screening methods on the basis of the reaction of the rice varieties to seedling blight. *Bangladesh J. Bot.* **14** (1): 56-61.

Akibode, S. and Maredia, M. (2011). Global and regional trends in production, trade and consumption of food legume crops. *Research Gate*. Department of Agricultural, Food and Resource Economics Michigan State University, Michigan, United States.

Anonymous, (1986). Annual report, 1985-86. Plant Pathology Division, Bangladesh Rice Research Institute (BARI), Gazipur, Bangladesh. pp. l9.

Asgari, M.R. and Mayee, C.D. (1991). Comparative efficacy of management practices on stem and pod of groundnut. *Indian Phytopathology*. **44**: 328-332.

BBS, (2009). Bangladesh Bureau of Statistics, Ministry of Planning, Government of the People's Republic of Bangladesh, Dhaka, Bangladesh. p. 63.

BBS, (2017). Bangladesh Bureau of Statistics. Ministry of Planning. Dhaka, Bangladesh. p.107.

BBS, (1995). Yearbook of Agricultural Statistics of Bangladesh. Bangladesh Bureau of Statistics, Ministry of Planning, Government of the People's Republic of Bangladesh, Dhaka. Bangladesh.

Bakhsh A., Ghafor, Zubeir M. and Iqbal S.M. (1991). Genotype International for grain yield in lentil, *Pakistan. J. Agril*. **12**: 102-105.

Bakr, M.A. (1994). Progress in pulse diseases management. In: Proc. Workshop on transfer of technology of CDP crops under Research Extension linkage program, held at BARI, Gazipur, Bangladesh, pp. 150-158.

Bakr, M.A., and Zahid, M.I. (1986). Stemphylium blight: a new foliar disease of lentil in Bangladesh. *Bangladesh Journal of Plant Pathology*. **2** (1): 69-70.

BARI Hand book, (2005). *Dal Fasol Utpadon Poddoti*, Bangladesh Rice Research Institute, Joydebpur, Gazipur, Bangladesh. pp. 101-104.

Bedard, T., Risula, D. and Olekson, A. (2010). Pulse Growers "Overview of the Canadian Pulse Industry 2009". Canada: Agriculture and Agri–Food Canada.

Begum, F. and Bhuiyan, M.K.A. (2007). Integrated control of seedling mortality of lentil caused by *Sclerotium rolfsii. Bangladesh J. Plant Pathol*. **23** (1& 2): 17-24.

Begum, F.M.S. (2003). Integrated control of seedling mortality of lentil caused by *Sclerotiuu rolfsii*. MS Thesis. Department of Plant Pathology, Bangabandhu Sheikh Mujibur Rahman Agricultural University, Gazipur, Dhaka, Bangladesh.

Clancey, B. (2009). World pulse outlook: Report to the Saskatchewan Pulse growers. Canada. p.17.

Dey, T.K., Saha A.K., Rahman, M. and Ali, M.S. (2004). Biocontrol potential of *Trichoderma spp*. against *Sclerotium rolfsii* causing stem and tuber rot of potato. *Bangladesh J. Plant Pathol*. **20**(1&2): 17-20.

Dey, T.K., Ayub, A., Ahmed, H.U. and Rahman, M.H. (1993). Screening of chickpea germplasm against Fusarium wilt. *Bangladesh J. Plant Pathol*. **9(**1&2): 29-30.

Dwivedi, S.K. and Ganesh Prasad, (2016). Integrated Management of *Sclerotium rolfsii*: An overview, *European Journal of Biomedical and Pharmaceutical*, **3**(11): 137-146.

Erskine, W., Sarker, A. and Ashraf, M. (2011). Reconstructing an ancient bottleneck of the movement of the lentil (*Lens culinaris*) into South Asia. *Genet. Res. Crop Evol*. **58**(3): 373-391.

Erskine, W., Witcombe, J.R, (1984). Lentil germplasm catalog, International Center for Agricultural Research in the Dry Areas (ICARDA), Aleppo, Syria. p. 363.

Erskine, W. (1997). Lessons for breeders from landraces of lentil. *Euphytica*. **93**(1): 107–112.

Elad, Y., Chet, I. and katan, J. (1980). *Trichoderma harzianum*: A bio-control agent effect against *Sclerotium rollsii* and *Rhizoctonia solani. Phytopathology*, **70**: 119-121.

Faruk, M.I., Rahman, M.L., Islam, M.N., Rahman , M.M., and Rahman M.A. (2004). Assessment of carrier materials to formulate *Trichoderma harzianum* bio-fungicide for controlling foot and root rot disease of brinjal in seed bed. *Agricultural Science*. **2**(2): 21-30.

Food and Agricultural Organization of the United Nations (FAO), (2014). FAO Statistical Database. USA: Food and Agricultural Organization of the United Nations.

Feedipedia, (2012). http://www.feedipedia.org/node/284. Retrieved on 12 February 2021.

Fried, M and Middelboe, V. (1977). Measurement of amount of nitrogen fixed by legume crop. *Plant and Soil*. **47**: 713-715.

Ganeshan, G. (1997). Fungicidal control of Sclerotium basal rot of cluster bean cv. Pusa Naubahar. *Indian Phytopathol*. **50**(4): 508-512.

Grichar, V.J. and Bosweel, T.E. (1987). Comparison of Lorsban and Tilt with Terrachlor for control of southern blight on peanut. The Texas. Agriculture Experiment Station. USA.

Garret, S.D. (1956). Biology of root infecting Fungi, Cambridge University Press, London. p. 293.

Gupta, S.C. and Kolte, S.J. (1982). A comparative study of isolates *Macrophomina phaseolina* from leaf and root of groundnut. *Indian Phytopathol.* **35**: 619-623.

Harlan, J.R. (1992). Crops and man, Second Ed. *American Society of Agronomy*, Madison, Wisconsin, USA. p. 284.

Harman, G.E, Howell C.R, Viterbo A., Chet I. and Lorito, M. (2004) *Trichoderma* species-opportunistic, avirulent plant symbionts. *Nat. Rev. Microbiol*. **2**: 43-56.

Hossain, M.D., Meah M.B., Siddique M.K., (1999) . Effect of Bavistin and Rhizobium on foot rot and root rot of lentil. *Bangladesh J. Plant Pathol*. **15**: 1-4.

Hoque, M.A., Hamim, I., Haque, M.R., Ali, M.A. and Ashrafuzzaman, M. (2014). Effect of some fungicides on foot and root rot of lentil. *Univ. J. Plant Sci*. **2**(2): 52-56.

Hoque, M.A., Hamim, I., Haque, M.R., Ali, M.A. and Ashrafuzzaman, M. (2012). Effect of some fungicides on foot and root rot of lentil. *Univ. J. Plant Sci*. **2**(2): 28-36.

ISTA. (1999). International Rules for Seed Testing. International Seed Testing Association, Seed Science and Technology, Zurich, Switzerland. pp. 333.

Jahan, N., Sultana, N., Zinat, S.R., Hossain, I. and Hossain, D.M. (2017). Biological control of Sclerotium rot of sugarbeet. *Bangladesh J. Plant Pathol*. **33**(1&2): 29-36.

Kandel, H. and Ashley, R. (2013). Growing Lentil in North Dakota. North Dekoda Agricultural Experiment station. Fargo, North Dakota 58108. http://www.ag.ndsu.edu/pubs/plantsci/rowcrops/a1636.pdf. Retrieved on 12 February 2021.

Katti, D.V.S., Bhargava, S.N. and D.N. Shukla. (1983). Some studies on the saprophytic survival of *Sclerotium rolfsii* in soil. *Z. Pflaslsrankh pflpath pllschute*. **90**: 382-387.

Kashem, M.A., Rafiiq, M.M.A., Mondal, M.S. Islam and Latif M.A. (2015). Effect of times and levels of inoculum of *Trichoderma* for controlling root rot and collar rot of lentil. *Legume Research*. **39**(1): 123-128.

Kashem, M.A., I. Hossain and M.K. Hasna (2011). Use of *Trichoderma* in biological control of foot and root rot of lentil *(Lens culinaris Medik*). *International Journal of Sustainable Crop Production***, 6**(1): 29-35.

Kochhar, S.L. (2009). Economic Botany in the Tropics. MacMillan India, 1981. Pp. 122.

Khare, M.V., Agrawal, S.C. and Jain, A.C. (1979). Diseases of lentil and their control. Technical bulletin. Jabalpur, Madhya Pradwsh, India: Jawaharlal Nehru Krisi Viswa Vidyalaya. (http://www.hrpub.org). Retrieved on 12 February 2021.

Khalequzzaman, K.M. (2016). Control of foot root rot of lentil by using different management tools. ABC J. Adv. Res. **5**(1): 35-42.

Krishi Projukti Hatboi, (2015). Dal Fosol. Bangladesh Agricultural Research Institute, (BARI), Joydebpur, Gazipur, Bangladesh. p. 81.

Kumar, S., Barpete, S. and Kumar, J. (2013). Global lentil production: constraints and strategies. SATSA Mukhapatra – Annual Technical Issue. **17**: 1-13.

Lingaraju, S.M.S. (1977). Studies on *Sclerotium rolfsii* Sacc. with respect to survival in soil. University of Agricultural Science, Bangalore, India.

Lewis, J.A. and Larkin R.P. (1997). Extruded granular formulation with biomass of bio-control *Gliocladium virens* and *Trichoderma spp*. to reduce damping spp. to reduce damping-off of eggplant caused by *Rhizoctonia solani* and saprophytic growth of the pathogen in soil-less mix. *Plant Disease.* **82** (5): 501-506.

Maiti, S. and Sen, C. (1984). Integrated biocontrol of *Sclerotium rolfsii* with nitrogenous fertilizers and *Trichoderma harzianum*. *Indian Journal of Agricultural Science*. **55**: 464-468.

Mannan, M.A., Hasan, M, Hossain, I. Rahman, and Ismail, M.A. (2012). Integrated management of foot and root rot of lentil using bio-control agents under field condition. *J. Microbial. Biotechnol*, **22** (7): 883-888.

Materne, M., Siddique, K.H.M, (2009). Agro–ecology and lentil crop adaption. The Lentil botany, Production and use. Europe: CAB International Ltd. pp. 47–63.

Mehrotra, R.D. and Aneja, K.R. (1990). An introduction of Mycology. Willey Eastern Ltd. New Delhi, India. MacMillan India Ltd., Daryaganj, New Delhi. p. 658.

Misgana Mitik,. (2017). Management of Root Rot Diseases of Cool Season Food Legumes Crops in Ethiopia. *Journal of Plant Sciences*. **5**(4): 104-109.

Nargund, V.B.S., Kulkarni and Hegde, R.K. (1984). Studies on foot and root rot of wheat caused by *Sclerotium rolfsii* sacc. In Karnataka. V. Effect of different organic amendments on *S. rolfsii* Sacc. Causing foot and root rot of wheat, *Current research*. **13**: 76-77.

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

Parvin, R.M.S. (2013). Management of foot and root rot disease of betel vine caused by *Sclerotium rolfsii*, MS Thesis. Department of Plant Pathology, Sher-e-Bangla Agricultural University, Dhaka, Bangladesh.

Pratibha Sharma, Prashant P., Jambhulkar and Manokaran Raja (2016). Management of groundnut stem and root rot complex by using *Trichoderma harzianum* Th3 at field level. *Biological and integrated control of plant pathogens*. **117**: 101-105.

Preatibha Sharm, Raja, M. and Shanmugam, V. (2014).Status of *Trichoderma* research in India: A review. Division of Plant Pathology, Indian Agricultural Research Institute, New Delhi. India. *Indian Phytopath*. **67** (1): 1-19.

Punja, Z.K, Smith, V.L., Campbell, C.L. and Jenkins, S.F. (1985). Sampling and extraction procedure to estimate numbers, spatial patterns and temporal distribution of sclerotia of *Sclerotium rolfsii* in soil. *Plant Dis.* **69**: 469- 474.

Rahman, M.L., Haque, M.S., Mukit, Alam, A.K.B. and Ali, S. (1994). Response of *Sclerotium rolfsii* to different fungicides. *Bangladesh J. Plant Pathol.* **10**(1&2): 35-36.

Sabaghpour, S.H., Safikhani , M., Sarker, A., Ghaffari, A. and Ketata, H. (2004). present status and future prospects of Lentil cultivation in India. In proceeding of 5th European conference on grain legumes. Dijon, France.

Sarker, A. and Kumar, S. (2011). Lentils in production and food systems in West Asia and Africa. International Center for Agricultural Research in the Dry Areas (ICARDA), Aleppo, Syria. *Grain Legumes*. **57**: 46–48.

Shahiduzzaman, Md. (2015). Efficacy of fungicides and botanicals in controlling foot and root of lentil. *Bangladesh J. Agril. Res*. **40**(4): 711-715.

Senanayake, L., Knievel, D.P., Steven, S.E. (1987). Nodulation and symbiotic nitrogen fixation of cowpea (*Vigna unguiculata* L.). *Plant Soil*. **99**: 435-439.

Saigan, S., Seraji, A. and Moghaddam, S.A. (2008). Identification and investigation on antagonistic effect of *Trichoderma* spp. on tea seedlings white foot and root rot (*Sclerotium rolfsii Sacc*.) *in vitro* condition. *Pakistan Journal of Biological Sciences: PJBS*. **11**(19): 2346-2350.

Sattar, M.A., Podder., A.R., Chandra, M.C. and Rahman, M. (1996). The most promising BNF technology for green legume production in Bangladesh. BNF Association, Dhaka, Bangladesh. pp.15-20.

Singh, J. and Tripathy, S. C. (1999). Mycoflora association with stored seeds of *Lens esculenta*. Herbal Pesticide Lab., Dept. of Botany, Gorakhpur University, Gorakhpur, India.

Singh, R.S., Singh, D. and Singh, H. V. (1997). Effect of fungal antagonists on the growth on chickpea plants and wilt by *Fusarium oxysporum* f. sp. *ciceri*. *Plant Dis. Res*. **12**(2):103-107.

Sreenivasaprasad, S. and Maribhushanarao, K. (1993). Efficacy of *Gliocladium varens* and *Trichordarma longribra-chiatum* as bio-control agents of lentil root and stem rot disease. *Int. J. Pest Mangt*. **39**:167-171.

Sultana, N., Hoque, S., Faruq, A.N., Bhuiyan, M.Z.R. and Islam, N. (2015). *In-vitro* evaluation of selected bio-control agents against foot and root rot pathogens of lentil. *Scholarly Agric. Sci*. **5**(1): 8-15.

Srivastava, S.N. and Tripathi, R.C. (1998). Management of sugarbeet seedling disease complex by combination of fungicides. *Indian Phytopathol*. **51**(1):75-77.

Saxena, D.R. and Saxena, M. (1993). Evaluation of chemicals against biological drying of lentil. *Bhartiya Krishi Anushandhan Patrika***.** **8**(1): 25-29.

Shahjahan, M.S. (2005). Soil amendments for control of foot and root rot of lentil. Department of Plant Pathology, Bangladesh Agricultural University, Bangladesh.

Taylor, P.W., Rubeena. Ford R. (2003). Construction of an intra–specific linkage map of lentil (*Lens culinaris* ssp. *culinaris*). *Theor. Appl. Genet*. **107**(5): 910–916.

Treggy, G. (1956). Study of physiological characters of strains of *Sclerotium rolfsii* isolated from tomatoes and chilies. *Ann. Sper. Agr. N. S*. **10** (5): 1553-7574.

Weidong, C., Hari, C., Sharma, F. and Muehlbauer, J. (2011). Compendium of Chickpea and Lentil Diseases and Pests. *The American Phytopathological Society*. P. 54.

Wever, G.F. (1931). Blight of carrots caused by *Sclerotium rolfsii* with geographic distribution and host range of the fungi. *Phytopathology*. **21**:103-109.

Zapata, F., Danso, S.K.A., Hardarson, G. and Fried. M. (1987). Nitrogen fixation and translocation in field-grown fababean. *Agronomy Journal*. **79**: 505-509.

CHAPTER 7

APPENDICES

Appendix I: Particulars of the Agro-ecological Zone of the Experimental site:

Agro-ecological region	Madhupur Tract (AEZ-28)
Land Type	Medium high land
General Soil Type	Non-calcareous dark gray floodplain soil
Topography	Up land
Soil series	Tejgaon
Drainage	Fairly good
Field level	Above flood level
Firmness	Compact to friable when dry
Soil p^H	4.47-5.55
Organic matter content	0.82

Appendix II: Monthly mean weather of the experimental site

Monthly mean of average temperature (°c), average Relative humidity (%), Rainfall and Pressure (mbar) from October/2019 to March/2020 are given bellow-

Year	Month	Average Temperature (^{0}C)	Relative Humidity (%)	Rainfall (mm)	Air Pressure (mbar)
2019	October	27.6	78	188	1010.1
	November	24.9	74	37	1011.5
	December	19.3	74	5	1015.2
2020	January	18.5	76	21	1014.7
	February	21.6	59	1	1014.5
	March	26.4	57	3	1010.7

Source: Bangladesh Meteorological Department (Climate Division), Agargaon, Sher-e-Bangla Nagar, Dhaka-1207.