# ECO-FRIENDLY MANAGEMENT OF FOOT AND ROOT ROT DISEASE OF LENTIL BY USING ORGANIC SOIL AMENDMENTS

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#### BY

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## CERTIFICATE

This is to certify that thesis entitled, "ECO-FRIENDLY MANAGEMENT OF FOOT AND ROOT ROT DISEASE OF LENTIL BY USING ORGANIC SOIL AMENDMENTS" submitted to the Department of Plant Pathology, 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 Rekhea Bhaumik, Registration No. 14-06161 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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#### ECO-FRIENDLY MANAGEMENT OF FOOT AND ROOT ROT DISEASE OF LENTIL BY USING ORGANIC SOIL AMENDMENTS

#### ABSTRACT

Lentil (Lens culinaris) is one of the oldest and most familiar food legumes in Bangladesh. Foot and root rot is very common disease in lentil. It causes seedling death at early stage resulting very poor plant stand which ultimately produces low yield. For eco-friendly management of the disease a field experiment was conducted at Central Farm of Sher-e-Bangla Agricultural University, Dhaka-1207, during October 2019 to May 2020. Susceptible lentil variety BARI Masur-1 was selected as planting material. The field experiment was laid out in randomized complete block design (RCBD) with three replications. Seven treatments were used as organic soil amendments viz.  $T_0$ = Control,  $T_1$ = Biofumigation with raddish leaf,  $T_2$ = Biochar,  $T_3$ = Vermicompost,  $T_4$ = Mushroom compost-I,  $T_5$ = Mushroom compost-II and  $T_6$ = Trichocompost. The causal organism was isolated from naturally infected root of lentil and identified as Fusarium oxysporum by observing morphological and cultural characteristics. All organic soil amendments were applied after final soil preparation. Data were recorded on disease incidence, plant height, number of branches per plant, pods per plant, yield per plant and yield per plot. The lowest disease incidence (3.11%) was recorded in Trichocompost (T<sub>6</sub>) at 60 DAS. The treatments also caused appreciable improvement of plant growth over control. The tallest plant (37.33 cm), maximum number of branches (28) per plant, maximum no. of pods (47.51), maximum seed weight per plant (0.67g), and the yield (564.99 g/plot) were also recorded from Trichocompost ( $T_6$ ) treated plots followed by Vermicompost ( $T_3$ ), Biochar  $(T_2)$  and Biofumigation with radish leaf  $(T_1)$ . In this study it was found that soil amendments with Trichocompost showed most effective in controlling foot and root rot disease incidence with increasing yield of lentil. Furthermore, studies have to be conducted using these organic soil amendments to unfold the potential of other organic amendments in the management of foot and root rot of lentil.

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## LIST OF ABBREVIATIONS AND SYMBOLS

%	=	Percentage
/	=	Per
@	=	At the rate
@	=	At the rate
°C	=	Degree Celsius
μg	=	Microgram
μm	=	Micrometer
AEZ	=	Agro-Ecological Zone
BARI	=	Bangladesh Agricultural Research Institute
BBS	=	Bangladesh Bureau of Statistics
cm	=	Centimeter
CV	=	Coefficient of Variation
DAE	=	Department of Agricultural Extension
DAI	=	Days After Inoculation
DAS	=	Days After Sowing
e.g.	=	exempli gratia (L), for example
et al.,	=	Et Alia (and other)
etc.	=	Etcetera (and so further)
FAO	=	Food and Agricultural Organization
g	=	Gram (s)
ha	=	Hectare
hrs	=	Hours
i.e.	=	id est (L), that is
J.	=	Journal
Kg	=	Kilogram (s)
L	=	Litre
LSD	=	Least Significant Difference
$m^2$	=	Meter square
mg	=	Miligram
ml	=	Milliliter
MS	=	Master of Science
No.	=	Number
PDA	=	Potato Dextrose Agar

P <sup>H</sup>	=	Potential Hydrogen
PP	=	Pertaining page (s)
PSI	=	Pounds per square inch
RCBD	=	Randomized Complete Block Design
SAU	=	Sher-e-Bangla Agricultural University
Sp.	=	Species
SRDI	=	Soil Resource Development Institute
USDA	=	United States Department of Agriculture
viz.	=	Videlicet (Namely)
WHO	=	World Health Organization

#### **CHAPTER I**

#### INTRODUCTION

Lentil (Lens culinaris L.) is among the oldest pre-eminent domestic pulse crops in the world and one of the first agricultural crops having been grown more than 8500 years ago in the Middle East. In Bangladesh, lentil ranks first position in respect of consumers' preference along with second most important pulse crop in terms of area and production (Uddin et al., 2013). Lentils play significant role in human and animal nutrition, as well as the maintenance and improvement of soil fertility and productivity, because they are critical components of agroecosystems all over the world due to their ability to fix atmospheric nitrogen into usable plant proteins. Lentils are part of the legume (pea) family, all of which grow in a symbiotic relationship with soil-dwelling bacteria. The bacteria take gaseous nitrogen from the air and feed this nitrogen to the plant; in exchange, the plant provides carbohydrates to the bacteria. This is why legumes are said to "fix" nitrogen when they are turned under for the next crop or used for compost. Lentil is also important in crop diversification in the cropping systems of Bangladesh. It can be grown in rotation to cereal crops with a potential of fixing free nitrogen reached up to 107kg ha-<sup>1</sup>(Abraham, 2015).

In the global lentil scenario, Bangladesh is the world's fourth-largest lentil producer and the greater part of world production comes from Canada and India, producing 58% combined of the world total. The major lentil-growing countries of the world are Canada, India, Turkey, Australia, USA, Nepal, China, and Ethiopia. Different kinds of pulses, such as lentil, mungbean, black gram, grass pea, chickpea, cowpea, field pea, and pigeon pea are grown in Bangladesh. Amid all Pulses, lentils have been gaining increasing attention for their nutritive value as human diet in Bangladesh. The per capita 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). People called lentil the powerhouse of nutrients by reason

of it are the protein-rich commodity that fulfills the protein requirement of the huge population of our country. Among all pulses, lentil contains 59% carbohydrate, 25% protein, and 0.7% fat (Afzal et al., 1999). It contains more protein than any other agricultural produce and is nearer to animal flesh in food value for which it is often called poor man's meat (Hajong et al., 2020). For these reasons, lentils have long been recognized as an inexpensive, excellent alternative to animal proteins, and are considered as a potential whole food source for people affected by micronutrient malnutrition (Thavarajah et al., 2009). Pulses cultivation covers 2.22% of the total cultivated land in Bangladesh (BBS, 2019). Among the pulses, lentil commonly known as "Masur" is a popular pulse crop in Bangladesh and occupied 40.23% cultivation of pulse crops getting first position (BBS, 2019). Production of lentils has long lagged behind domestic demand in Bangladesh. Lentil is cultivated on 3,49,000 acre of land in 2019-2020 in our country (BBS, 2020). Farmers produce 1.75 lakh tons of lentil yearly but the yearly requirement of our country is around 6 to 7 lakh tons. Bangladesh has to import a huge quantity of pulses especially lentil for the reasons of increased consumption and inadequate domestic production by spending a lot of foreign exchange. Bangladesh imported from abroad about 5,08,850 metric tons in the year 2020. Bangladesh has favorable production environments such as weather conditions, soil fertility, water availability, and comparatively cheaper labor supply which can boost production in Bangladesh. The production of lentils is decreasing every year due to cultural and genetic factors, susceptibility to disease, poor management practices, poor planting materials, and delay in sowing by the farmers. Low yield potential, susceptibility to diseases, delayed sowing, drought, and weed infestation are the main production constraints to the lentil crop in Bangladesh (Uddin et. al., 2013).

Diseases are an important yield-limiting factor in lentil production. Lentil plants are vulnerable to a number of diseases caused by fungus, bacteria, and viruses. The productivity of lentil is reduced by pathogens through infection and damage to leaves, stems, roots, and pods (Hoque *et al.*, 2014). Fungal diseases of lentils

are the most important biological constraint to productivity (Taylor et al., 2007). The most common fungal diseases of lentils are foot and root rot, collar rot, alternaria blight, stem rot, grey mold, powdery mildew, rust, stemphylium blight, anthracnose, which are caused by Fusarium oxysporum, Sclerotium rolfsii, Rhizoctonia solani, Botrytis fabae, Erysiphe polygoni, Uromycis fabae, Peranospora lentis, Colletotrichum truncatum, respectively. Out of all the fungal diseases foot and root rot of lentil is a killer disease of lentil. These diseases can cause yield loss up 50% in farmer's fields (Tiwari et al., 2018). Foot rot (caused by Fusarium oxysporum and Sclerotium rolfsii) is considered the most significant and ruinous disease of pulses in almost all legume-growing countries of the world as well as in Bangladesh. Fusarium oxysporum and Sclerotium rolfsii are soil-borne as well as seed-borne pathogens commonly occur in legume-growing countries in the tropic and sub-tropic regions of the world. In Bangladesh, about 44% of lentil plants are infected by foot and root rot disease (Anonymous, 1986). It causes seedling death at an early stage resulting in very poor plant stand which ultimately produces very low yield (Hoque et al., 2014). But both of these pathogens attack the lentil plants from seedling to maturity stages but they are more destructive at the seedling stage. Foot and root rot diseases may cause 100% early seedling mortality in monoculture under conducive weather or resulting in very poor plant stand which ultimately produces very low yield. The fungi are soil-borne in nature so that organic soil amendment, botanicals, and biocontrol agents might be effective to control foot and root rot disease to increase the yield of lentil. The use of organic amendments reduces the risk of environmental pollution, health hazards and is not much cost to the growers. These types of research work are needed in Bangladesh among the farmers to create awareness about eco-friendly disease management. A very few investigations has so far been conducted in the discipline of controlling foot and root rot of lentil by organic soil amendments in Bangladesh. Kashem et al., (2011) reported that, for the control of foot and root rot in lentil, isolate of Trichoderma harzianum can inhibit the growth of Fusarium oxysporum in the in-vitro condition as well as field condition. Tricho-compost and Tricho-leachate was found most effective in controlling soil-borne diseases of cabbage (Naher *et al.*, 2012). Biofumigation by *Brassica spp.* are considered an effective and environmentally safe method for the control of soil-borne pests, including phytopathogens and weeds (Kapoor, 2013; Smolinska *et al.*, 2003). After decomposing of *Brassica* tissues, the volatile biocidal compounds namely isothiocyanates is released which help suppress soil borne pathogens. Application of biochar into the soil improves soil health by adding carbon to the soil and increasing plant growth by suppressing soil-borne pathogens (Egamberdieva *et al.*, 2016). Vermicompost has a tremendous potential to protect plants from diseases and its application to plants can coat leaf surfaces and reduce available sites for pathogen infection or increases microbial diversity that can kill harmful pathogens (Yatoo *et al.*, 2021). Spent mushroom compost mixed with solarized soil might have effective biocontrol potentiality for the suppression of *Fusarium oxysporum* sp. *lycopersi* on tomato plants (Salim *et al.*, 2017).

Considering the above facts the present study was undertaken to evaluate the efficiency of the organic soil amendment for controlling foot and root rot of lentil in field conditions of Bangladesh.

Thus, the experiment was undertaken keeping in mind to find out the effect of different soil amendments to assess the application of organic soil amendments on lentil production viz. biofumigation, botanicals, biocontrol agents against foot and root rot of lentil.

Considering the above-stated circumstances, points, and based on the available literature, this research work was designed to obtain the specific following objectives:

- 1. To isolate, identify and characterize the causal organism of foot and root rot disease of lentil; and
- 2. To evaluate some organic soil amendments in controlling foot and root rot disease of lentil.

#### **CHAPTER II**

#### **REVIEW OF LITERATURE**

Pulses are the major key food component along with the cereal in Bangladesh. Lentil is the second major pulse crop of Bangladesh in respect of acreage and production. It is cultivated as sole and intercrops. Lentil is one of the oldest domesticated and most appreciated food crops that has been grown in the world in terms of food, feed, and farming systems. Pulses constitute an integral part of the daily balanced diet as a direct source of protein for human beings in Bangladesh. 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 (*Dey et al.*, 1993; Anon., 1986).

#### 2.1. Importance of Lentil

Lentil is one of the oldest domesticated food legume crops in the world, playing a significant role in human and animal nutrition as well as soil fertility maintenance. Lentil is currently an important pulse crop grown in Bangladesh. It is a nutritious food legume and cultivated for its seed. The primary product of lentil is its seed, which has relatively higher contents of protein, carbohydrate, and calories compared to other legumes. It is the most desired pulse crop because of its high average protein content and fast cooking characteristic in many lentilproducing regions.

Khalequzzaman (2016) mentioned that being 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 can be used as a main dish, side dish, or in salads. Its flour is used to make soups, stews, purees, and mixed with cereals to make bread and cakes; and as food for infants (Abraham, 2015).

Garkoti *et al.* (2013) stated that pulses are valued for their high protein content and protein quality and thus, supplement the cereal based diets. The protein content of most pulses ranges from 17-24 per cent which is almost 2-3 times more than that found in cereals.

According to Kochhar (2009) lentil is also used for human consumption as a protein source in a diverse range of products and is an excellent source of vitamin A and provides fiber, potassium, B vitamins, and iron.

Sattar *et al.* (1996) reported that lentils (*Lens culinaris*) have been gaining increasing attention for their nutritive value as human diet. Lentil covers 40% of the production. It is a cheap source of protein for human beings and also for animals in our country.

#### 2.2. Fungal diseases of Lentil

Das *et al.* (2019) reported that the legume crop such as lentil suffers from a number of diseases which are caused by many fungi, bacteria, viruses, nematodes and plant parasites as a result decline the grain yield abruptly. The foot and rot root disease caused by *Rhizoctonia solani*, collar rot caused by *Sclerotium rolfsii*, Fusarium wilt caused by *Fusarium oxysporum* f. sp. *lentis* and damping off or seedling mortality caused by all those of pathogens are the main limiting factors to successful cultivation.

According to Kumar *et al.* (2013) lentil plant can suffer substantial yield losses from various biotic and abiotic stresses.

Taylor *et al.* (2007) stated that fungal diseases of lentils are the most important biological constraint to productivity. *Ascochyta lentis* (ascochyta blight) and *Fusarium oxysporum* f. sp. *lentis* (fusarium wilt) are the major fungal pathogens that can cause severe yield losses in most lentil growing regions of the world.

Lentil suffers from an attack of a number of seed-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*, *Uromyces fabae*, *Erysiphe polygoni*, *and Peronospora lentis*, respectively (Singh and Tripathy, 1999; Khare *et al.*, 1979).

#### 2.3. Foot and root rot disease of lentil

The soil-borne pathogens *Fusarium oxysporum* and *Sclerotium rolfsii* commonly occur in the tropics and sub-tropics of the world causing foot and root rot of many crops (Aycock, 1966).

Bhalla *et al.* (1992) reported that among all the diseases of lentil, Fusarium wilt caused by *Fusarium oxysporum* f. sp. *lentis* is the most important biological constraints to productivity of lentil worldwide.

#### 2.4. Significance of foot and root rot disease of lentil

Kaur *et al.* (2018) stated that *Fusarium osysporum* is a vascular, seed borne and soil borne pathogen. In favorable conditions high temperature with drought, wilting can look within 3–4 weeks after sowing. In susceptible genotypes plant death rate goes up to 80% at flowering and pod formation stage due to disease.

Khalequzzaman (2016) reported that the foot and root rot fungi of lentil can attack the crop during any time from seedling to flowering stage and are comparatively more destructive at the seedling stage.

In a long-term favorable environment, foot and root rot of can cause up to 100% yield loss (Kumar *et al.*, 2010).

Begum and Bhuiyan (2007) observed that amid all diseases of lentil, foot and root rot was the serious one along with, it may cause 100% morality of seedlings in the field under monoculture and conducive weather conditions.

According to Agrios (2005) that soil-borne plant pathogens causing root rot and damping-off disease are among the limiting factors in plant production all over the world. These pathogens cause economic yield losses on faba bean (*Vicia fabae*), Lentil (*Lens culinaris*), and pea (*Pisum sativum*), and control is rather difficult.

According to Dey *et al.* (1993), the foot and root rot of lentil caused by *Fusarium oxysporum* and *Sclerotium rolfsii* are common in Bangladesh. This root rot fungus leads to economic losses in a variety of crop plants, including lentil.

Begum and Bhuiyan (2007) and Fakir (1983) reported that seedling diseases are more destructive and causing 30-40% yield loss in lentil.

In Bangladesh about 44% lentil plants are infected by foot and root rot disease (Anonymous, 1986).

Lentil is the cheap source of protein for human beings and also for animals in Bangladesh (Sattar *et al.*, 1996).

#### 2.4. Symptoms of foot and root rot disease

*Fusarium oxysporum* is a soil borne, root pathogen colonizing the xylem vessels and blocking them completely to cause wilting.

Jiskani *et al.* (2021) stated that the *Fusarium* attacked crops showing stunting, wilting, withering, chlorosis, necrosis, and defoliation of plant parts which conclusively results in the death of the whole plant.

Mitiku (2017) mentioned that after the seedling has emerged, continued cool wet weather often results in root rot. Symptoms will include stunted, yellow plants and may be mistaken for nitrogen. When the plant is dug up, the roots will be much thinner than a healthy plant or there may be no secondary roots at all. Roots will be discolored, and the color and pattern of discoloration depend on the pathogen infecting the roots.

Pouralibaba *et al.* (2017) declared that the symptoms of wilting in the field include wilting of old leaves, stunting of plant development, and leaf atrophy and curling of the lower part of the plant moving up to the stem of the infected plant. Plants eventually turn yellow and die.

Hoque *et al.* (2014) observed that foot and root rot of lentil can cause seedling death at an early stage, resulting in a very poor plant stand which ultimately produces a very low yield.

Garkoti *et al.* (2013) reported that the disease appears in the field in patches at both seedling and adult stages. Seedling wilt is characterized by sudden drooping, followed by drying of leaves and seedling death and adult wilt symptoms appear from flowering to late pod-filling stage and are characterized by sudden drooping of top leaflets of the affected plant, leaflet closure without premature shedding, dull green foliage followed by wilting of the whole plant or individual branches.

Ignjatov *et al.* (2012) found that the infection usually occurs in the form of chlorosis, leaf wilting and browning of the vascular system and the cross-section of the stem leads to vascular necrosis but the roots look healthy, proliferation

and nodulation are reduced, and usually, the vascular system does not have external discoloration.

Stoilova and Chavdarov (2006) asserted that *Fusarium oxysporum* wilt occurs in the field, which is patchy and occurs in the seedling stage or adult reproductive stage of crops.

According to Madhavi *et al.* (2006) that the initial symptom of Fusarium wilt is usually the golden yellow of a single leaflet or twig or a slight wilting and drooping of the lower leaves on a single stem in gardens and fields.

Under field conditions, the symptoms produced by *F. oxysporum* f. sp. *lentis* include stunting, marked reduction of the root system, internal vascular discoloration of the lower stem, and wilting (Tosi and Cappelli, 2001).

De Cal *et al.* (2000) reported that at an advanced stage of infection with *Fusarium oxysporum*, browning of the vascular system can be viewed, and the pathogen induces severe wilting of plants via blocking off xylem vascular bundles and impeding the movement of water.

#### 2.5. Causal organism of foot and root rot disease

The foot and root rot of lentil is mainly caused by *Sclerotium rolfsii* and *Fusarium oxysporum*.

Garkoti *et al.* (2013) mentioned that *Fusarium oxysporum* is soil borne fungus, which can survive in the soil and plant debris in the absence of its host for a period of 3-4 years. The disease is favoured by low soil temperature, 30 percent soil water holding capacity and increasing plant maturity. Yield losses depend on the stage at which the plant wilts; it can be 100 percent when wilt occurs at pre pod stage, about 67 percent when it occurs at the pre harvest stage.

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

#### 2.6. Isolation and identification of *Fusarium oxysporum*

Kripalini *et al.* (2018) analyzed fourteen isolates of *Fusarium oxysporum* f. sp. *pisi* from different districts of Manipur was studied for its cultural, morphological, and pathogenic variability. The mycelia colour varied from white to light pink, purple and pale yellow colour. The radial growth of the isolates ranged from 5.4 to 8.9cm at 8 days after inoculation at  $26\pm10$  <sup>o</sup>C in 90 mm petriplates. Sporulation of all isolates showed moderate to profuse. The size of microconidia ranged from 11.6 x 3.1 to 25.2 x 6.2 µm and the size of microconidia ranged from 3.02 x 2.1 µm to 9.2 x 5.6 µm. The number of septation of macroconidia most of the isolates in microconidia is mostly sickle-shaped and microconidia most of the isolates are oval-shaped. The conidial colours of all the isolates were found hyaline.

Patra and Biswas (2017) studied eleven isolates of *F. oxysporum* f. sp. *ciceri* for its cultural, morphological, and pathogenic variability. The radial growth of isolates ranged from 72 mm to 87 mm at seven days after inoculation on PDA medium. Sporulation of isolates was profuse to moderate. The size of macroconidia was ranged from 13-15 x 2-3  $\mu$ m to 15-19 x 3-4  $\mu$ m, in micro-conidia was from 3-4 x 1-2  $\mu$ m to 5-6 x 2-3  $\mu$ m. The number of septa in macro-conidia was mostly 2-3 and micro-conidia are mostly no septum and some are 0-1. Conidia are hyaline. The shape of most macro-conidia is the sickle shape and micro-conidia are round to oval.

Teixeira *et al.* (2017) reported that the Colony color may vary from white, pink, purple, and violet. The average macroconidia length varied from 19.8 to 61.0  $\mu$ m and width varied from 3.15 to 6.0  $\mu$ m, respectively, among the isolates. Microconidia length varied from 5.80 to 8.05  $\mu$ m.

Bayona *et al.* (2011) identified *Fusarium oxysporum* isolates under a microscope and found that it produces aerial mycelium with a strong light to dark purple pigment diffusing into agar, visible from both sides of the Petri dish. It produces microconidia, which were oval or kidney-shaped, single or two-celled. They were produced on false heads with short monophialides. Intercalar chlamydospores were abundant, mostly single but sometimes in pairs. Macroconidia were slightly curved, thin walled, generally three septae, with a foot-shaped basal cell.

By studying characteristics of *Fusarium oxysporum* f. sp. *passiflorae* isolates, Dariva (2011) reported that *Fusarium solani* forms cylindrical macroconidia, with no convex curvature, which as seen in *Fusarium oxysporum* f. sp. *passiflorae*.

Dariva (2011) also recorded by studying the characteristics of *Fusarium oxysporum* f. sp. *passiflorae* isolates, an average mycelial growth of 6.36 cm in PDA after four days of incubation and colony color vary from white, cream, and violet in different tinges at the seventh day of incubation.

Ciampi *et al.* (2009) isolated two isolates of *Fusarium* species from wilted callas. The isolates were identified as *F. solani* (20%) and *F. oxysporum* (80%). They mentioned that the colony color in PDA medium is light and dark violet, salmoncolored, purplish brown, all with cottony mycelium, without exudate. The microconidias of *F. solani* do not have septa and originate from long conidiophores, while those of *F. oxysporum* have one or two septa and originate form short conidiophores. *F. oxysporum* produces *a*bundant, sickle-shaped, 3 to 5 septa with distinctive cellular foot (27-55 x  $3-5 \mu m$ ).

Leslie and Summerell (2008) identified *Fusarium oxysporum*. Mycelia may be floccose, sparse or abundant and range in color from white to pale violet. Abundant pale orange or pale violet macroconidia are produced in a central spore mass in some isolates. Small pale brown, blue to blue-black or violet sclerotia

may be produced abundantly by some isolates. *F. oxysporum* usually produces a pale to dark violet or dark magenta pigment in the PDA but some isolates produce no pigment at all. Some isolates of *F. oxysporum* mutate readily to the pionnotal form or to a flat "wet" mycelial colony with a yellow to orange appearance when cultured on PDA. Most isolates produce abundant pale orange sporodochia and macro conidia on it. Macroconidia are short to medium length, straight to slightly curved, relatively slender and thin walled, usually 3- septate. Microconidia are oval to elliptical or kidney shaped, aseptate, abundant in aerial mycelia. Chlamydospores are usually formed singly or in pairs, but also may be found in clusters or in short chains. May be either terminal or intercalary in aerial, submerged, or surface hyphae.

Booth (1977) identified *Fusarium oxysporum* and found that microconidia of *F*. *oxysporum* varied in shape, from elliptical to cylindrical, with 0 to 2 septa.

Booth (1971) reported that lentil wilt is caused by *Fusarium oxysporum* f. sp. *lentis*. The fungus is septate, profusely branched growth on PDA at 25°C, initially white turning light buff or deep brown later and fluffy or submerged. The growth becomes felted or wrinkled in old cultures. Various types of pigmentation (yellow, brown, whitish/cream, crimson dark purple, light orange) may be observed in culture on solid medium. Micro conidia may be usually borne on simple and short conidiophores, which arise laterally on the hyphae. They are oval to cylindrical, straight or curved and measure  $2.5 - 3.5 \times 5 - 11 \mu m$ . Macro conidia are borne on branched conidiophores, thin walled, 1 to 6 septate, fusoid, pointed at both end and measures  $3.5 - 4.5 \times 25 - 65 \mu m$ . Chlamydospores are formed in old cultures, which are smooth or rough walled, terminal intercalary and may be formed singly, or in pairs or in chains.

Chattopadhyay and Sengupta (1967) proposed that *F. oxysporum* var. *lentis* on lentil should be named as *F. oxysporum* f. sp. *lentis*.

# 2.7. Management foot and root rot disease of lentil by applying organic soil amendments

According to Iqbal *et al.* (2019), foot and root rot disease can be controlled by many physical as well as biological methods that have a low cost of production and are safe for the environment.

Harman (2011) and Singh *et al.* (2011) conducted that numerous studies have shown that biological control offers an environmentally friendly alternative to protect plants from soil-borne pathogens.

#### **Reviews on Tricho-compost**

Mukhopadhyay and Kumar (2020) reported that the species of *Trichoderma* uses several mechanisms to control the growth and proliferation of harmful pathogens such as parasitism, competition and antibiosis.

Faruk (2019) investigated the effect of tricho-compost for the management of barley seedling disease caused by *Sclerotium rolfsii* through soil amendment in the field of Plant Pathology Division, Bangladesh Agricultural Research Institute (BARI), Gazipur, Bangladesh during 2013-14, 2014-15 and 2015-16. The yield of barley was increased over control due to the trichocompost 41.09, 38.55 and 44.03% in the year of 2013-14, 2014-15 and 2015-16, respectively.

Das *et al.* (2019) used poultry refuge mixed with bio-agent i.e *Trichoderma harzianum* isolate Pb-7 is compatible and has combined effect in controlling the pathogenic fungi viz. *Rhizoctonia solani*, *Fusarium oxysporum* f. sp. *lentis* and *Sclerotium rolfsii*. They observed post-emergence mortality of plants due to damping off, foot and root rot, collar rot and wilt disease of lentil variety in BARI- Masur-6 were significantly reduced after the field treatment with *T. harzianum* fortified compost. The highest shoot length and root length also recorded for *T. harzianum* fortified compost treated plot.

Akter *et al.* (2018) conducted an experiment to assess the antagonism of *T. harzianum* against *F. oxysporum* by *in-vitro* and *ex-vitro* to reduce lentil wilt. Tricho-compost treated lentil plants shown maximum reduction of wilt incidence compared to *T. harzianum* spore suspension and other combinations in *in-vitro* pot study. Wilt disease appeared most (63%) in combining treatment of conventional compost + Fusarium spore suspension. Population density of both test fungi in conventional compost is high and maximum population of *T. harzianum* is found in conventional compost + Tricho-compost application.

Khalequzzaman (2016) conducted a research in Pulse Research Centre, Ishurdi, Pabna to figure out the effect of chemical, botanical and biocontrol agent against the foot and root rot disease of lentil. The foot and root rot fungi are comparatively more destructive at the seedling stage and it can attack the crop any time during its growth stages and this pathogenic fungi are soil-borne in nature; hence, seed treatment with chemical, botanicals and biocontrol agents might be effective to control foot and root rot disease and to increase yield of lentil.

Faruk and Rahman (2016) conduct a survey on management of stem and root rot disease of lentil through soil amendment with Tricho-Compost in two different AEZ's of Bangladesh Agricultural Research Institute (BARI), Gazipur and Pulses Research Station, Madaripur of Bangladesh. Among six treatments, Trichocompost was found more efficient in the reduction of seedling mortality and acceleration of plant growth with increased grain yield of lentil under *S. rolfsii* and *F. oxysporum* inoculated pot culture as well as field experiments. The yield of lentil was sharply increased over control due to the *T. harzianum* formulations and Provax in both locations. In BARI, Gazipur in the year of 2012-13 and 2013-14 yield increase over control in case of Trichocompost were 25.68 & 47.35%, similarly in Madaripur, yield increase over control were 17.14 & 38.67%.

Shahiduzzaman (2015) carried out an experiment at Regional Pulses Research Station of Bangladesh Agricultural Research Institute (BARI), Madaripur, in the cropping seasons of 2011-12 and 2012-13. All treatments with fungicides as well as botanicals increased the crop yield significantly over control and the highest yield (37.82%) was achieved with Provax 200WP followed by Tricho compost (30.10%) and Bavistin 50WP (23.89%). The study also found that trichocompost can increase yield of lentil over control which is ranges from 29.24 to 30.10%.

Faruq *et al.* (2014) carried out a field experiment at Sher-e-Bangla Agricultural University farm during 2011-2012 cropping season to test the efficacy of *Trichoderma harzianum*  $T_{22}$  and some selected soil amendments viz. poultry waste, coco-dust, vermi-compost, ash, saw-dust, khudepana (*Azolla pinnata*), cowdung and solarized sand against Fusarium wilt disease of eggplant. At 65 DAS, the highest (43.33%) wilt incidence was found in control plot and lowest (10%) incidence in *Trichoderma harzianum* used plot in terms of suppressing wilt incidence, increasing plant growth and fruit yield. Poultry waste and Vermi-compost also showed promising performance against the disease. Poultry waste treated plot showed best growth performance and gave highest yield followed by Vermi-compost, *Trichoderma harzianum* and coco-dust treated plot.

Kashem *et al.* (2011) evaluated 14 isolates of *Trichoderma* spp. for the control of foot and root rot of lentil caused by *Fusarium oxysporum*. The study showed that the isolate of *T. harzianum* can be used to control foot and root rot disease of lentil in Bangladesh because it showed the lowest foot and root rot incidence (6.9%), the highest seed germination (82.08%), maximum plant stand (93.12%), and highest seed yield (3726.67 kg/ha). They concluded that the isolate TG-2 of *T. harzianum* can be used to control foot and root rot disease of lentil in Bangladesh and also stated that the biological management of soil-borne diseases is increasingly gaining stature as a possible practical and safe approach.

Harman *et al.* (2004) conducted research on *Trichoderma* as a virulent symbionts and the disease suppression through *Trichoderma* is based on hyperparasitism, antibiosis, induced resistance in the host plant and competition for nutrients and space.

Tjamos *et al.* (1992) ovserved that, the majority of *Trichoderma spp.* are potential antagonist of plant pathogenic fungi and have been broadly used as the most important biocontrol agent.

Papavizas (1985) conduncted an investigation on several strains of *Trichoderma* and *Gilocladium* as the potential bio control agent and found that *Trichoderma* spp. might be effective as biocontrol agents of various soil-borne plant pathogenic fungi such as *Fusarium, Pythium, Rhizoctonia* and *Sclerotium*.

#### **Reviews on Biofumigation**

Srivastava and Ghatak (2017) reported that biofumigant crops act as break crops and disrupts the lifecycle of pests and diseases. The disease Suppression may result from direct biocidal toxicity and suggested that 50 ton per hectare fresh biomass is required to maximize the pathogen suppression.

Kareem and Matar (2016) conducted an experiment to evaluate the biofumigation with crushed radish leaves and seed meals to manage the disease complex caused by *Fusarium oxysporum* and *Meloidogyne* spp. in eggplants under greenhouse conditions. *F. oxysporum* and *Meloidogyne* spp. as proved by the high significant reduction of the disease complex incidence and severity that cause, compared with the control. The antagonistic effect of radish material may come from the compounds that released in the soil during covering with the plastic sheets.

Kapoor (2013) carried out an experiment on soil solarization as an eco-friendly method for famers in agriculture and reported that biofumigation by *Brassica* 

spp. are considered effective and environmentally safe methods for the control of soil-borne pests, including phytopathogens and weeds. Biofumigation effect brassica cover crops have great potential to improve soil quality in organic systems (Yodar, 2014).

Matthiessen and Shackleton (2005) investigated biofumigation and environmental impacts on the biological activity of diverse pure and plantderived isothiocyanates and stated that, biofumigation by *Brassica* spp. is an alternate method for plant disease suppression. *Brassica* spp. contain different kinds of natural biocidal compounds, such as glucosinolate.

Smolinska *et al.* (2003) investigated the isothiocyanates produced by *Brassicaceae* species as the inhibitor of *Fusarium oxysporum* and reported that, after decomposing of *Brassica* tissues, the volatile biocidal compounds (mainly isothiocyanates) can be released which helps in reduction of pathogenic population of *Fusarium oxysporum* by inhibiting mycelial growth completely suppressed conidial and chlamydospore germination of all isolates.

Alternative measures for disease control including biofumigation that may offer a strategy ecofriendly to manage soil borne fungal pathogens (Subbarao *et al.*, 1999; Ramirez-Villapudua and Munnecke, 1988).

#### **Reviews on Vermicompost**

Tian *et al.* (2021) conducted a pot experiment in American ginseng to investigate the effects of vermicompost (VF), biochar (BF), and a combination of vermicompost and biochar (VBF) applied after soil sterilization on the incidence of Fusarium root rot. The highest bacterial richness and diversity were observed in the rhizosphere soil of VBF. Besides, VF and VBF significantly increased the relative abundance of beneficial bacteria (*Pseudomonas, Lysobacter*, and *Chryseolinea*) in the rhizosphere soil. Zhao *et al.* (2019) conducted an experiment on the effects of three organic amendments (rice straw, chicken manure compost, and vermicompost) on the suppression of *Fusarium oxysporum* f. sp. *lycopersici* (*Fol*) in soil. Vermicompost was found the most effective organic fertilizer to suppress *Fol* in long-term continuous tomato cropping soil. Vermicompost directly introduces beneficial bacteria to the soil and improves the soil environment, which results in the propagation of the beneficial bacteria and the empirical evidence consistently suggests that bacteria belonging to the genera *Nocardioides*, *Ilumatobacter* and *Gaiella* represent the keystone microbial taxa in the inhibition of *Fol*.

Szczech (1999) conducted a series of experiment on suppressiveness of vermicompost against Fusarium wilt of tomato and the infection of tomato plants by *Fusarium oxysporum* was lower in all of the tested potting mixtures that were amended with Vermicompost. Vermicompost also stimulated plant growth in infested and infested treatments.

#### **Reviews on Biochar**

Biochar, a carbon-rich by-product of thermo chemical conversion, is now considered as potential soil ameliorant worldwide. It has received immense global importance as an organic amendment in the field of sustainable agricultural waste management for soil improvement, C accretion and environmental management (Janardhan and Krishna, 2021).

According to Jaiswal *et al.* (2018) that adding biochar to soil and soilless media can help protect plants against diseases caused by soilborne pathogens. There are a number of direct and indirect mechanisms that are potentially responsible for this effect.

Rogovska *et al.* (2017) conducted glasshouse and field experiments to determine the effect of biochar on severity of soybean root disease caused by *Fusarium*  *virguliforme*. Both pot and field experiment showed that biochar can significantly reduce the effect of soybean root disease caused by *Fusarium virguliforme*.

Egamberdieva *et al.* (2016) reported that the application of biochar to soil is considered to have the potential for long-term soil carbon sequestration, as well as for improving plant growth and suppressing soil pathogens.

Elmer and Pignatello (2011) evaluated the effect of Biochar as soil amendments on mycorrhizal associations and Fusarium crown and root rot of asparagus in replant soils. In greenhouse studies, biochar added at 1.5 and 3.0% (wt/wt) to asparagus field soil caused proportional increases in root weights and linear reductions in the percentage of root lesions caused by *Fusarium oxysporum* f. sp. asparagi.

Zwart and Kim (2012) asserted that soil amendment with biochar is thought to confer multiple benefits to plants including induction of systemic resistance to plant pathogens.

Fidanza *et al.* (2010) reported that, after mushroom production, the substrate is removed and pasteurized with steam heat to eliminate the potential for unwanted fungi and weed seeds, and then it can be used as compost. Fresh mushroom compost was previously called spent mushroom substrate or "mushroom soil". Although the aged mushroom compost has been used as an organic fertilizer and soil amendment for plant production in agriculture.

#### **CHAPTER III**

#### **MATERIALS AND METHODS**

#### **3.1. Experimental site**

The whole experiment was divided into two main parts. One is a Field experiment and another is a Laboratory experiment. The laboratory experiment was conducted in Dr. M A Wazed Miah Research Centre, Post Graduate laboratory, Department of Plant Pathology, Sher-e-Bangla Agricultural University, and the field experiment was conducted at the Central Farm of Sher-e-Bangla Agricultural University, Dhaka-1207. The coordinates for the field experimental area were 23°75″ N latitude and 90°35″ E longitude with an elevation of 8.3 meters from sea level.

#### **3.2.** Duration of the experiment

The experiment was carried out during the Rabi season from October 2019 to April 2020 and laboratory research was done in between December 2019 to march 2020. The seeds of lentil were sown on 26<sup>th</sup> November, 2019 and harvested on 24th March, 2020.

#### **3.3. Selection and collection of planting materials**

Seeds of lentil (*Lens culinaris*) variety, BARI Masur-I (Utfala) were selected for both study based upon susceptibility to foot and root rot disease of Lentil. This variety was released in 1991 and seed was collected from Pulses wing, Bangladesh Agricultural Research Institute (BARI), Joydevpur, Gazipur -1701.

#### **3.4.** Laboratory experiment

#### 3.4.1. Materials used in experiment

The following materials were used in the lab experiment -

- Small tools viz., scissor, inoculation needles, knife, blade, scale, forceps, cork borer, sprit lamp, electronic balance, wrapping tape, blotter paper, aluminium foil.
- 2) **Large equipments** viz., incubator, autoclave, laminar air flow or inoculation chamber, oven, refrigerator, microscope.
- 3) **Chemicals** viz., 70% ethanol, hexisol, agar powder, dextrose powder, lactic acid, glycerine, cotton blue.
- 4) **Glassware** viz., Petri dish, Funnel, Test tube, Beaker, Conical flask, Slides, and Cover slip.
- 5) Data recording materials viz., microscope, computer, camera.
- Others viz., zip lock polythene bag, cotton cloth, potato, tissue paper, Vermicompost, Tricho-Compost, Biochar, Mushroom Composts, Raddish Leaf etc.

## **3.4.2.** Isolation and identification of the causal organism of foot and root rot of disease lentil

#### **3.4.2.1.** Collection of disease sample

Diseased lentil seedlings showing typical symptoms of foot and root rot were collected from the Central Farm of SAU and kept in zip lock polyethylene bags to maintain the moist condition. Then the samples were carried to the Dr. M A Wazed Miah Research Centre, Department of Plant Pathology, MS Laboratory, placed at Sher-e-Bangla Agricultural University to study the visual symptoms and isolation of the pathogen.

## **3.4.2.2.** Isolation of pathogenic fungi by growing on moist blotter paper (Incubation Method)

The fresh collected diseased plant root samples showing typical disease symptoms were cut into small pieces having rotted along with healthy portion, washed thoroughly in running tap water, and kept in a sterile Petri dish. Then, the cut sample pieces were surface sterilized with 70% Ethanol for 30 seconds Hereafter, the cut pieces were transferred to a sterile dish containing sterile water and washed thoroughly with two to three changes of sterile water to free from chemical. Three-layer water-soaked blotting paper was placed into the sterile petri dish for making a moist chamber. Two small pieces of the sample were placed aseptically on the moist blotting paper and covered with lid and kept at room temperature for incubation. The petri dishes were observed daily.



Plate 1. Part of disease sample on moist blotting paper

#### 3.4.2.3. Preparation of culture medium

Potato dextrose agar is a general-purpose basal medium used for the identification, cultivation, purification, enumeration, and preservation of fungi. It also aids in cultivating and differentiating pathogenic and non-pathogenic fungi. Potato Dextrose Agar, often denoted as PDA media was used. Nutritionally, potato dextrose agar contains dextrose as a carbohydrate source which serves as a growth stimulant and potato infusion that encourages luxuriant growth and sporulation of most fungi and pigment production in some cases.

Agar is added as the solidifying agent. A specified amount of sterile lactic acid to be incorporated to lower the pH of the medium to 3.5 so that bacterial growth is inhibited. PDA media was prepared according to standard procedure is given below-

Two hundred gram of peeled potato slices were taken in a sauce pan containing 1 liter of water and boiled for 30 minutes and after boiling filter through cheesecloth, saving effluent, and kept in a conical flask. Then weighing all the ingredients taken in a conical flask (Composition in Appendix V). Thoroughly, mixed all ingredients by using a glass rod and a few amounts of water were added to this solution to make the volume 1L. Boil to dissolve completely. Then the mouth of the conical flask was closed by a cotton plug and wrapped with aluminum foil paper. Sterilize media by autoclaving at 121°C temperature for 15 minutes at 15 PSI. The media was looked like light amber-colored clear to slightly opaque gel. Before dispensing the media in petri dishes 1 ml acetic acid was added to the media under laminar airflow cabinet and adjusted. Aseptically dispense into sterile petri dishes.

#### 3.4.2.4. Purification and maintenance of pure culture

When the pathogen was grown in a moist chamber, then a bit of mycelium was taken with the help of a sterilized needle and transferred to PDA plates aseptically to obtain the culture of that pathogen. Then, the plates were incubated in an inverted fashion in an incubator at 27 °C temperature. Profuse mycelial growth of a fungus on the plates was observed after incubation. Then, the pathogen was isolated, identified, and recorded. Conclusively, it was repeatedly sub-cultured on potato dextrose agar (PDA) medium to get a pure culture of the organism.

#### 3.4.2.5. Slide preparation and identification of the causal organism

A slide was prepared from the pure culture of the causal organism and observed under the compound microscope.

#### **3.4.3.** Storage and preservation of pure culture

When the pure culture of targeted fungus was achieved, this pathogenic isolate was stored in PDA slant for further studies. For this, 5 mm culture discs of the fungal mycelium were cut with the help of sterilized cork borer and transferred aseptically in potato dextrose agar slants and allowed to grow. The pure culture slants were sealed with paraffin wax and stored in a refrigerator at 4 °C.

#### **3.5. Field experiment**

#### **3.5.1.** Characteristics of experimental site Soil

The soil of the experimental site belongs to the agro-ecological regions of Madhupur Tract" under AEZ No. 28 (Appendix I). 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 area was flat, having available irrigation and drainage system. The land was above flood level and sufficient sunshine was available during the experimental period. Soil samples from 0-15 cm depths were collected from the experimental field. The analytical data of the soil sample collected from the experimental area was determined by Soil Resources and Development Institute (SRDI), Dhaka. The physicochemical properties of the soil are presented in Appendix II. The pH of the soil was 4.47 to 5.55 and organic carbon contents is 0.82.

#### **3.5.2.** Climatic condition of experimental site

The experiment was conducted in the winter season (Rabi) of Bangladesh. The experimental plot is under the sub-tropical climate zone which is characterized

by comparatively scanty rainfall, low humidity, low temperature, relatively short days during Rabi season(October to March), and high rainfall, high humidity, high temperature, and long day period during Kharif season(April to September). The monthly mean for daily maximum, minimum, and average temperature, relative humidity (RH%), and monthly total rainfall received at the experimental field during the period of the experiment had been collected from Bangladesh Meteorological Department, Agargaon, Dhaka (Appendix III).

#### **3.5.3. Land preparation**

A medium high land was chosen with good drainage system for the field experiment. The land was prepared mechanically in 8<sup>th</sup> November, 2019. The field was first ploughed on 10<sup>th</sup> November 2019. Crop residue, weeds, rocks were removed from the soil. The clods of the land were hammered to make the soil into small pieces. The soil was prepared into good tilth followed by laddering. The soil of the field was leveled before seed sowing. The final ploughing and land preparation was done on 15<sup>th</sup> November, 2019.

#### 3.5.4. Application of fertilizers and manures

According to standard recommendation doses, all fertilizers and manures were applied during land preparation for the cultivation of BARI Masur –1 (Krishi Projukti Hatboi, 2019).

Fertilizer/Manures	Recommended Dose (Kg/ha)				
Urea	40-45				
TSP	80-90				
МОР	30-40				
Cowdung	5000-6000				
Gypsum	50-55				
Boron	7-10				

**Table 1.** Recommended doses of fertilizer and manure in the field is given below:

(Source: Krishi Projukti Hatboi, BARI, Joydebpur, Gazipur, 2019)

#### 3.5.5. Design of the experiment

The experiment was laid out in Randomized Complete Block Design (RCBD) with three replications. The total field was divided into three blocks (replication). Each block was divided into seven experimental units. The size of each experimental unit was  $2m \times 1.7m$ . The treatments were assigned randomly at each block.

#### 3.5.6. Layout preparation

The field layout was done as per experimental design 25th October, 2019. The unit plot size was  $2m \times 1.7m$  and plot to plot distance was 1m and block to block distance was 1 m (Appendix- IV).

#### 3.5.7. Collection of soil amendments

Soil amendments are generally used to improve plant growth conditions. Vermicompost and mushroom compost-1 were collected from SAU FAB LAB. Mushroom compost -2 was collected from Mushroom Development Institute, DAE, Savar. Biochar were collected from Cristian Commission for Development in Bangladesh (CCDB), and Manikganj. For biofumigation (with Raddish leaf), organically grown raddish plant in seedbed of SAU Central Farm is used. Trichocompost was collected from local nursery in Dhaka city.

#### **3.5.8.** Application of treatments

There were six organic soil amendment were used as treatment with one control. The treatments were as follows-

 $T_{0}= \text{Control}$   $T_{1}= \text{Biofumigation with raddish leaf @ 50 ton fresh weight /ha}$   $T_{2}= \text{Biochar @ 4-5 ton/ha}$   $T_{3}= \text{Vermicompost @ 5-6 ton /ha}$   $T_{4}= \text{Mushroom compost-I @ 5-6 ton /ha}$   $T_{5}= \text{Mushroom compost-II @ 5-6 ton /ha}$ 

 $T_6$ = Trichocompost @ 2-2.5 ton/ha

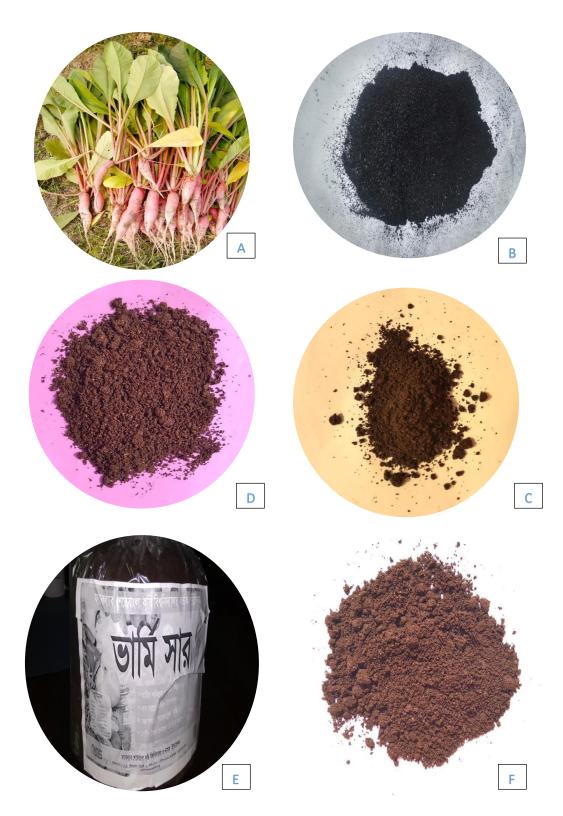


Plate 2. Different organic soil amendments used for experiments; A. Raddish leaf B. Biochar C. Mushroom compost-I D. Mushroom compost-II E. Vermicompost F. Trichocompost

#### Application of Biofumigation into the experimental plots

Radish plants were grown on the seedbed of Central Farm of SAU (Plate 3A). At 25 DAS, the plants were harvested (Plate 3B) and finely chopped into small pieces and incorporated into the unit plot soil @ 5kg/m<sup>2</sup>. After this, the plot was covered by polythene sheets until they decomposed in order to maintain the moist environment which may speed up the decomposition process (Plate 3D). Then we checked 7 days interval and apply little water if needed and cover it with polythene. After 21 days when it was fully decomposed the sheets were removed and the soil was sun-dried before seed sowing.





Plate 3. Growing radish plants in seedbed and application of Biofumigation into the field

#### 3.5.9. Seed rate and seed sowing

Before seed sowing, lines were made and seeds were sown in furrows at broadcast method. The total amount of seeds are taken and divided for each unit plot and kept in separate polybags. Seed rate was 35-40 kg/ha (Hoque, 2014). Immediately, the furrows were covered with the soil by hand after sowing.

#### **3.5.10. Intercultural operation**

Seed germination was started after 4-5 days after sowing. Different intercultural operations were done after sowing like thinning, weeding, irrigation with the aim to achieve normal hygienic conditions for the growth of crop.

#### 3.5.11. Irrigation and drainage /Water management

Light irrigation was given after sowing and continued throughout the whole cultivation process as per necessity. Excess water was removed immediately to save the crop from stagnant water.

#### 3.5.12. Weeding

Weeding was performed several times, during the growth of the crop with intention of keeping the plants free from completion for nutrients and water.

#### 3.5.13. Thinning

Extra plants were removed at 20 DAS after sowing by keeping healthy plants in line to maintain an optimum population.

#### **3.5.14.** Tagging of plants

Data collection was done based on counting diseased plants randomly as well as healthy plants and thereafter measuring the height (cm), pod number etc.

After harvesting the dry matter accompanied with yield, based on their treatment identity was recorded.

#### **3.5.15.** Disease of the plants

Natural infection of the plants was considered in this experiment.

#### 3.5.16. Data collection

During the field experiment, the Growth and physiological parameters of the different stages were recorded. The yield parameters were recorded at harvest. In each unit plot, five plants were chosen randomly and then data were documented on the following parameter-

#### **Disease related parameters**

1) Percent disease incidence per plot

#### **Growth parameters**

- 1) Plant height (cm)
- 2) No. of branch/plant

#### Yield and yield contributing factors

- 1) No. of pods per plant
- 2) Seed/yield per plant
- 3) Yield (g/plot)

#### 3.5.17. Procedure of data record

#### **3.5.17.1.** Data recording on growth and yield contributing characters

#### 3.5.17.1.1. Disease incidence

The self-observation was done for investigating the total number of infected plants per plot at different growth stages. The disease incidence of foot and root rot of lentil was recorded at 15 days intervals by choosing any 5 plants randomly in zig zag pattern against the entire plants that existed at that instant of inspection in the plot was expressed in percentage.

Along these lines, the percent plant incidence of lentil per plot was calculated by using the following formula:

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

#### **3.5.17.2.** Data recording on growth parameters

#### **3.5.17.2.1. Plant height (cm)**

Plant height was quantified from randomly chosen five sample plants from the ground level up to the tip of the leaf in centimeter (cm) by using a meter scale at a different stage of plant growth and thereafter the average height of five plants was considered as the final height per plant in centimeter for each plot.

#### 3.5.17.2.2. Number of branches per plant

Randomly 5 plants were taken from each plot and the number of branches per plant was counted. The average branch number of five plants was taken for the final branch number per plant.

#### 3.5.17.3. Harvesting of crops

When the plants in the experimental field showed 80 to 90% maturity based on straw color, pod filling, pod color, etc. indices. At maturity, total plants per plot were harvested and tagged based on plot identity. Total 21 plots were separately harvested for data collection.

#### 3.5.17.3.1. Number of pods per plant

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

#### **3.5.17.3.2.** Yield (g/plant)

The yield was recorded by weighing the total harvested pod per plant and measured in gram for final data preparation.

#### 3.5.17.3.3. Yield (g/plot)

The yield was recorded by weighing the total harvested pod per plot and measured in gram for final data preparation.

#### 3.6. Percent increase in yield over control

Percent increase in yield over control was calculated by using the following formula-

Percent increase in yield over control = 
$$\frac{T-C}{C} \times 100$$

Where,

C = Yield in Control

T = Yield in Treatment

#### 3.7. Statistical analysis of data

The recorded data for various characters were analyzed statistically. The accumulated data from different parameters were compiled, tabulated, and subjected in Microsoft Excel 2019. Analysis of variance (ANOVA) and LSD test at 5% level of significance was done by using the computer package program Statistix 10.0 to figure out the significant difference among the treatment means.

#### **CHAPTER IV**

#### **RESULTS AND DISCUSSION**

This chapter depicts the after effect of different organic soil amendments on the growth and yield of lentil. Data collected from the experiment on various observations were recorded, tabulated, statistically analyzed and illustrated through figures with appropriate headings to describe the findings. The analyses of variance (ANOVA) of growth and yield parameters are presented in Appendix (VIII-XI).

#### 4.1. Symptoms of foot and root rot disease of lentil

Lentil is grown as a high protein crop worldwide. Foot and root rots are an important yield-limiting factor in lentil. *Fusarium* species are most frequently identified as causal agents of root rot of lentil. The organisms that cause the disease are seed or soil-borne and can infect the plant at the seedling stage, along with the later stage of development.

The initial typical symptoms produced by *Fusarium* sp. are drooping, yellowing, wilting, and dying of the lower leaves, regularly on one part of the plant (Plate 4). These symptoms may be found successively on young leaves, with one or more branches being affected and others enduring healthy. Latterly, a few weeks, dying of whole plant (Plate 4). The diseased plant root is thinner than healthy plant and did not have hairs due to infection by the pathogen and tap root is discolored, and elapsed (Plate 5). These disease appears in the field in patches at both seedling and adult reproductive stages of lentil (Plate 6).

These symptoms were similar to the symptoms studied by Jiskani *et al.*, 2021; Mitiku, 2017; Pouralibaba *et al.*, 2017; Hoque *et al.*, 2014; Garkoti *et al.*, 2013; Stoilova and Chavdarov, 2006. According to Jiskani *et al.* (2021) *Fusarium* attacked crops showing stunting, wilting, withering, chlorosis, necrosis and defoliation of plant parts which conclusively results in the death of the whole plant. Mitiku (2017) reported that after the seedling has emerged, if cool wet weather continued often results in root rot. Symptoms will include stunted, yellow plants and may be mistaken for nitrogen. When the plant is dug up, the roots will be much thinner than a healthy plant or there may be no secondary roots at all. Stoilova and Chavdarov (2006) reported that Fusarium oxysporum occurs wilt in the field, which is patchy and occurs in the seedling stage or adult reproductive stage of crops. Pouralibaba et al. (2017) declared that the symptoms of wilting in the field include wilting of old leaves, stunting of plant development, and curling of the lower part of the plant moving up to the stem of the infected plant. Plants eventually turn yellow and die. Hoque et al. (2014) observed that foot and root rot of lentil can cause seedling death at an early stage, resulting in a very poor plant stand which ultimately produces a very low yield. Garkoti et al. (2013) found that the disease appears in the field in patches at both seedling and adult stages. Seedling wilt is characterized by sudden drooping, followed by drying of leaves and seedling death and adult wilt symptoms appear from flowering to late pod-filling stage and are characterized by sudden drooping of top leaflets of the affected plant.



Plate 4. Infected plants showing foot and root rot with brownish at the leaves and stem along with healthy plants in the field



Plate 5. Infected plant showing foot and root rot symptoms



Plate 6. Disease appears in patches at adult stages in the field

#### 4.2. Identification of the causal organism of foot and root rot of lentil

Isolation of the causal organism was made from lentil seedling roots showing typical symptoms of the disease. The fungus was successfully isolated on PDA medium and obtained profuse mycelial mat and maximum sporulation. The fungus was identified as *Fusarium oxysporum*. In pure culture, the fungal colony was initially pale white to light purple (7B), cottony with profuse aerial mycelium which gradually turned violet in color (Plate 7C). Aged culture appeared completely dark violet in color with no aerial mycelium. Mycelium of Fusarium is thin, septate and branched and hyaline to tinge pink in color at maturity. Its conidiophoreis short, slender, branched, and bears conidia terminally and which is known as sporodochium (a fruiting structure that have a cluster of conidiophores woven together on a mass of hyphae). Fusarium sp. mainly produces two types of asexual spores namely, macro conidia and micro conidia. Macroconidia are produced in a sporodochium (Plate 8D), which is an erumpent crowded cluster of conidiophores arising from stroma to form a cushion-like mass that supports the macroconidia. Macro conidia are hyaline, large multicellular usually two to several celled, fusiform (bent at the both pointed ends) to sickle-shaped, with an elongated apical cell and pedicellate basal cell (Plate 8F). Microconidia are produced on the aerial mycelium singly or in short chain. Micro conidia are one or two-celled, hyaline, smaller than macroconidia, round to oval or curved in shape (Plate 8D, 8E).

Similar kinds of results have been documented by several researchers (Kriplani *et al.*, 2018; Teixeira *et al.*, 2017; Bayona *et al.*, 2011; Dariva, 2011; Ciampi *et al.*, 2009). Kripalini *et al.* (2018) analyzed fourteen isolates of *Fusarium oxysporum* for its cultural, morphological, and pathogenic variability. The mycelia colour varied from white to light pink, purple. Sporulation of all isolates showed moderate to profuse. The number of septation of macroconidia was mostly 2-3 whereas in microconidia most of the isolates were found with no septum. The shape of the macroconidia is mostly sickle-shaped and microconidia

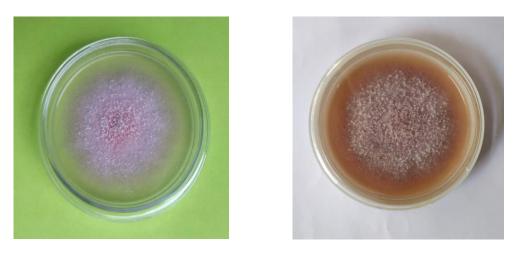
most of the isolates are oval-shaped. The conidial colours of all the isolates were found hyaline. Patra and Biswas (2017) investigated eleven isolates of F. oxysporum for its cultural, morphological, and pathogenic variability. Sporulation of isolates on PDA medium was profuse to moderate. The number of septa in macro-conidia (sickle shaped) was mostly 2-3 and micro-conidia (round to oval) are mostly no septum and some are 0-1. Conidia are hyaline. The colony color may vary from white, pink, purple, and violet (Teixeira et al., 2017). The average macroconidia length varied from 19.8 to 61.0 µm and width varied from 3.15 to 6.0 µm, respectively, among the isolates. Microconidia length varied from 5.80 to 8.05 µm. According to Bayona et al. (2011), Fusarium oxysporum isolaletes produced aerial mycelium with a strong light to dark purple pigment diffusing into agar, visible from both sides of the petri dish. It produced microconidia, which were oval or kidney shaped, single or two-celled. They were produced on false heads with short monophialides. Macroconidia were slightly curved, thin walled, generally three septae, with a foot-shaped basal cell. Ciampi et al. (2009) studied two isolates of Fusarium and asserted that the colony color in PDA medium is Light and dark violet, with cottony mycelium, without exudate. The microconidias of F. oxysporum have one or two septa and originate form short conidiophores. F. oxysporum produced macroconidia which is abundant, sickle-shaped, 3 to 5 septa.



(A) **3DAI** 







(C) 12 DAI

(D) 25 DAI

Plate 7. Growth of *Fusarium oxysporum* at different DAI (Days after inoculation).

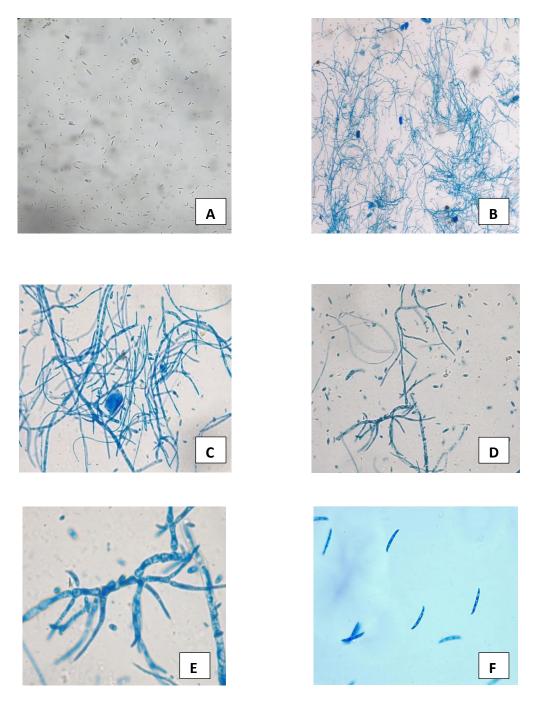


Plate 8. Different microscopic view of *Fusarium oxysporum*. (A) Young macro and microconidia of *F. oxysporum* at 10x (without staining) (B) Mycelia of *F. oxysporum* with microconidia at 4x (stained with cotton blue) (C) Mycelia of *F. oxysporum* with microconidia at 10x (stained with cotton blue) (D) Branched mycelium, branched septate conidiophore with macro and micro conidia (stained in cotton blue at 10x) (E) Branched sporodochia with micro conidia at 40x with staining (E) Sporodochial fusiform macro-conidia stained with cotton blue at 40x.

### 4.3. Effects of different organic soil amendments on disease incidence of foot and root rot of lentil at 15, 30, 45 and 60 DAS

On the basis of visible typical symptoms, the effect of different organic soil amendments on disease incidence of foot and root rot of lentil at 15 days after sowing (DAS) during growth period had been recorded and is presented in Table 2. Seven different treatments were compared with each other for disease incidence recorded at 15 DAS, 30 DAS, 45 DAS and 60 DAS.

At 15 DAS, the lowest (4.61%) disease incidence was recorded from treatment  $T_6$  (Trichocompost), followed by  $T_3$  (Vermicompost) 6.51%,  $T_2$  (Biochar) 8.73%,  $T_1$  (Biofumigation with raddish leaf) 11.31%, and  $T_4$  (Mushroom compost-I) 12.09%, whereas treatment  $T_0$  (Control) showed the highest disease incidence (32.26%), followed by,  $T_5$  (Mushroom compost -II) 16.25%.

At 30 DAS, the highest disease incidence (40.96%) was recorded in  $T_0$  (Control) followed by  $T_4$  (Mushroom compost-I) 19.69 %, and  $T_5$  (Mushroom compost - II) 12.20%. The lowest disease incidence (6.11%) was recorded in  $T_6$  (Trichocompost) followed by  $T_3$  (Vermicompost) 8.29%,  $T_2$  (Biochar) 8.72 %, and  $T_1$  (Biofumigation with raddish leaf) 10.14%.

At 45 DAS, the highest disease incidence (45.03%) was recorded in  $T_0$ . The lowest (4.23%) disease incidence was recorded in  $T_6$  followed by  $T_3$  (Vermicompost) 5.93%,  $T_2$  (Biochar) 7.70 %,  $T_1$  (Biofumigation with raddish leaf) 9.01 %,  $T_4$  (Mushroom compost-I) 17.92%,  $T_5$  (Mushroom compost-II) 16.06%.

Finally, at 60 DAS, least (3.11%) disease incidence was recorded in T<sub>6</sub> followed by T<sub>3</sub> (Vermicompost) 3.35%, T<sub>1</sub> (Biofumigation with raddish leaf) 6.67%, T<sub>4</sub> (Mushroom compost-I) 9.53%, T<sub>5</sub> (Mushroom compost-II) 13.02%. The highest (50.74%) disease incidence was recorded in T<sub>0</sub> (Control).

These findings are partially supported by Shahiduzzaman, 2015 and Kashem *et al.*, 2011. Kashem *et al.* (2011) evaluated 14 isolates of *Trichoderma* spp. for the

control of foot and root rot of lentil caused by *Fusarium oxysporum*. The study showed that the isolate of *T. harzianum* can be used to control foot and root rot disease of lentil in Bangladesh because it showed the lowest foot and root rot incidence (6.9%), the highest seed germination (82.08%), maximum plant stand (93.12%). Shahiduzzaman (2015) carried out an experiment at Regional Pulses Research Station (BARI), Madaripur. Seven treatments along with fungicides as well as botanicals increased the crop yield significantly over control and the highest disease reduction was achieved with Bavistin 50WP and Tricho-compost was similar. The study also found that trichocompost can increase yield of lentil over control which is ranges from 29.24 to 30.10%.

Table 2. Effect of different organic soil amendments on disease incidence of
foot and root rot of lentil at 15, 30, 45 and 60 DAS

	Disease Incidence (%)				
Treatments	15 DAS	30 DAS	45 DAS	60DAS	
T <sub>0</sub> =Control	32.26 a	40.96 a	45.03 a	50.74 a	
T <sub>1</sub> =Biofumigation with raddish leaf	11.31 c	10.14 d	9.01 d	6.76 d	
T <sub>2</sub> =Biochar	8.73 d	8.72 d	7.70 de	4.81 e	
T <sub>3</sub> =Vermicompost	6.51 e	8.29 de	5.93 ef	3.35 f	
T <sub>4</sub> =Mushroom compost-I	12.09 c	19.69 b	17.92 b	9.53 c	
T <sub>5</sub> =Mushroom compost-II	16.25 b	13.20 c	16.06 c	13.02 b	
T <sub>6</sub> =Trichocompost	4.61 f	6.11 e	4.23 f	3.11 f	
CV (%)	7.37	8.82	6.68	5.99	

\*Values in a column with same letter (s) do not differ significantly (p=0.05)

## 4.4. Effect on different organic soil amendment on growth and growth contributing parameters of lentil

Effects of different organic soil amendments on the basis of different growth and growth contributing parameters viz. plant height (cm), No. of branch was recorded at 45, 60 and 75 DAS and data was presented in Table 3.

#### a) Plant height (cm)

Plant height was recorded for seven treatments compared with each other at 45, 60 and 75 DAS during the growth period and presented in Table 3.

At 45 DAS, the highest (21.10cm) plant height was recorded in  $T_6$  (Trichocompost), followed by  $T_2$  (Vermicompost) 17.71 cm,  $T_1$  (Biofumigation with raddish leaf) 16.74cm,  $T_4$  (Mushroom compost-I) 16.73 cm,  $T_2$  (Biochar) 13.11cm, and  $T_5$  (Mushroom compost-II) 15.78cm, whereas lowest plant height was recorded in  $T_0$  (Control) 7.99cm.

At 60 DAS, the lowest (13.81cm) plant height was recorded in  $T_0$  (Control) followed by  $T_4$  (Mushroom compost-I) 16.73 cm and highest (29.34cm) plant height was recorded in  $T_6$  (Trichocompost), followed by  $T_3$  (Vermicompost) 25.93 cm,  $T_2$  (Biochar) 24.53 cm,  $T_1$  (Biofumigation with raddish leaf) 23.08 cm, and  $T_5$  (Mushroom compost-II) 22.24 cm.

At 75 DAS, the highest (37.33 cm) was recorded in  $T_6$  (Trichocompost) followed by  $T_3$  (Vermicompost) 30.60 cm,  $T_2$  (Biochar) 29.49 cm,  $T_1$  (Biofumigation with raddish leaf) 27.86 cm,  $T_4$  (Mushroom compost-I) 27.01 cm, and  $T_5$  (Mushroom compost-II) 24.13 cm and lowest (16.25 cm) plant height in  $T_0$ .

#### b) No. of branch

At 45 DAS, maximum (3.72) number of branch was recorded in  $T_6$  (Trichocompost), followed by  $T_3$  (Vermicompost) 3.2,  $T_1$  (Biofumigation with raddish leaf) 2.85,  $T_2$  (Biochar) 2.83,  $T_4$  (Mushroom compost-I) 2.49 and  $T_5$ 

(Mushroom compost-II) 2.16 and  $T_0$  (Control) shown the minimum (0.96) no. of branch.

At 60 DAS, minimum (1.69) no. of branch is recorded in  $T_0$  (Control) followed by and  $T_5$  (Mushroom compost-II) 3.03, whereas maximum (5.81) number of branch was recorded in  $T_6$  (Trichocompost), and second maximum no. of branch was recorded in  $T_3$  (Vermicompost) 4.65, followed by  $T_2$  (Biochar) 4.24,  $T_1$ (Biofumigation with raddish leaf) 3.92,  $T_4$  (Mushroom compost-I) 3.35.

At 75 DAS, maximum (6.20) no. of branch was recorded in  $T_6$  (Trichocompost) followed by  $T_3$  (Vermicompost) 5.85,  $T_1$  (Biofumigation with raddish leaf) 4.75,  $T_2$  (Biochar) 4.47,  $T_4$  (Mushroom compost-I) 4.24, and  $T_5$  (Mushroom compost-II) 3.92 and minimum (1.95) no. of branch was in  $T_0$  (Control).

	Plant height(cm) at different		No. of branch at different days				
		days					
Treatments	45 DAS	<b>60 DAS</b>	75 DAS	45 DAS	60 DAS	<b>75 DAS</b>	
T <sub>0</sub> =Control	7.99 d	13.81 d	16.25 f	0.96 f	1.69 e	1.95 d	
T <sub>1</sub> =Biofumigatio	16.74	23.08 cd	27.86	2.85 c	3.92 c	4.75 b	
n with raddish	bc		cd				
leaf							
T <sub>2</sub> =Biochar	16.11	24.53 bc	29.86	2.83 c	4.24 bc	4.47 bc	
	bc		bc				
T <sub>3</sub> =Vermicompo	17.71 b	25.93 b	30.60 b	3.20 b	4.65 b	5.58 a	
st							
T <sub>4</sub> = Mushroom	16.73	20.62 c	27.00 d	2.49 d	3.35 d	4.24 bc	
compost-I	bc						
T <sub>5</sub> =Mushroom	15.78 c	22.24 de	24.13 e	2.16 e	3.03 d	3.92 c	
compost-II							
T <sub>6</sub> =Trichocompo	21.10 a	29.34 a	37.33 a	3.72 a	5.81 a	6.20 a	
st							
CV (%)	5.71	5.19	4.91	6.76	6.50	8.75	

 Table 3. Effect of different organic soil amendment on growth and growth contributing parameters of lentil

\*Values in a column with same letter (s) do not differ significantly (p=0.05)

# 4.5. Effect of different organic soil amendment on yield and yield contributing parameters of lentil

Effect of different organic soil amendments on yield and yield contributing characters of lentil viz. no. of pods per plant, yield/ plant (g), yield/plot (g) were evaluated and tabulated in Table 4.

#### a) No. of pods per plant

Among various treatments of organic soil amendments, the maximum number of pod (47.51) were recorded in Tricocompost treated plots  $T_6$  followed by ( $T_3$ ) and ( $T_2$ ) and the minimum number of pods (19.61) were recorded in control plots  $T_0$ .

#### b) Yield (g/plant)

The highest (0.67g) yield/plant was found on  $T_6$  (Trichocompost), followed by  $T_3$  (Vermicompost),  $T_2$  (Biochar),  $T_1$  (Biofumigation with raddish leaf) were 0.64g, 0.63g. Treatment  $T_6$ ,  $T_3$  and  $T_2$  were statistically similar in terms of yield per plant. The lowest yield/plant was recorded on  $T_0$  (Control) 0.18 g.

#### c) Yield (g/plot)

The highest 564.99g yield per plot was recorded in case of  $T_6$  where Tricocompost was applied as soil amendment. Treatment  $T_3$  (Vermicompost) was produced the second highest yield (506.40 g) followed by treatment  $T_2$ (Biochar) and  $T_1$  (Biofumigation using raddish leaf) yielding 497.74 g and 439.99 g per plot, respectively. Treatment  $T_3$  and  $T_2$  were statistically similar in terms of yield. The lowest yield per plot (297.63 g) was recorded in  $T_0$  (Control).

Treatments	No. of pods/plant	Yield/plant (g)	Yield/plot (g)
T <sub>0</sub> =Control	19.61 e	0.18 d	297.63 e
$T_1$ =Biofumigation with raddish leaf	36.10 c	0.55 b	439.99 c
T <sub>2</sub> =Biochar	42.22 b	0.63 a	497.74 b
T <sub>3</sub> =Vermicompost	43.89 ab	0.64 a	506.40 b
T <sub>4</sub> =Mushroom compost-I	27.59 d	0.44 c	406.92 cd
T <sub>5</sub> =Mushroom compost-II	27.99 d	0.41 c	401.77 d
T <sub>6</sub> =Trichocompost	47.51 a	0.67 a	564.99 a
CV (%)	5.96	7.72	4.53

Table 4. Effect of different organic soil amendments on yield and yieldcontributing characters of lentil in field

\*Values in a column with same letter (s) do not differ significantly (p=0.05)

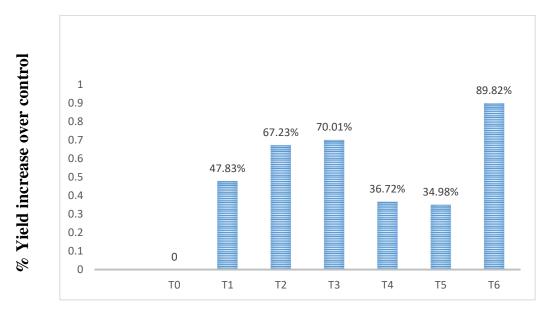
### 4.6. Effect of different organic soil amendments on percent yield increased over control against *Fusarium oxysporum* causing foot and root rot of lentil

Effect of organic soil amendments on percent yield increased over control against *Fusarium oxysporum* causing foot and root rot of lentil was evaluated under field condition and shown in Figure 2.

The maximum (89.82%) yield increased over control was found in case of the treatment  $T_6$  (Trichocompost) followed by  $T_3$  (Vermicompost),  $T_2$  (Biochar) and  $T_1$  (Biofumigation with raddish leaf) resulting 70.01, 67.23 and 47.83%, respectively. The lowest yield increased was observed in  $T_4$  (Mushroom compost-II) 36.72% preceded by  $T_5$  (Mushroom compost-I) 34.98 %.

The findings of this studies were also observed by several researchers (Faruk, 2019; Faruk and Rahman, 2016, Shahiduzzaman, 2015). Faruk (2019)

investigated the effect of Tricho-compost for the management of Barley Seedling Disease Caused by Sclerotium rolfsii through soil amendment in the field of Plant Pathology Division, Bangladesh Agricultural Research Institute (BARI), Gazipur, Bangladesh during 2013-14, 2014-15 and 2015-16. The yield of barley was increased over control due to the Trichocompost 41.09, 38.55 and 44.03% in the year of 2013-14, 2014-15 and 2015-16, respectively. A very few works have been done on the management of plant disease with bio control in Bangladesh (Das et al., 2019). Faruk and Rahman (2016) studied the efficacy of Trichoderma harzianum based Tricho-inocula, Tricho-compost and seed treating chemical fungicide Provax against foot and root rot diseases of lentil caused by Sclerotium rolfsii and Fusarium oxysporum in two different agro-ecological zones of Bangladesh Agricultural Research Institute (BARI), Gazipur and Pulses Research Station, Madaripur of Bangladesh. The yield of lentil was sharply increased over control due to the T. harzianum formulations and Provax in both locations. In BARI, Gazipur in the year of 2012-13 and 2013-14 yield increase over control in case of Ttrichocompost were 25.68 and 47.35%, similarly in Madaripur, yield increase over control were 17.14 and 38.67%. Shahiduzzaman (2015) conducted an experiment to find out the efficacy of few fungicides along with botanicals in controlling foot and root rot of lentil at Regional Pulses Research Station of Bangladesh Agricultural Research Institute (BARI), Madaripur, in the cropping seasons of 2011-12 and 2012-13. The study showed that Trichocompost can increase yield of lentil over control which is ranges from 29.24 to 30.10%.



#### Treatments

### Figure 1. Effect of different organic soil amendments on percent yield increased over control against *Fusarium oxysporum* causing foot and root rot of lentil

Here,

 $T_0 = Control$ 

 $T_1$ = Biofumigation with raddish leaf @ 50 ton fresh weight /ha

T<sub>2</sub>= Biochar @4-5 ton/ha

T<sub>3</sub>= Vermicompost @ 5-6 ton /ha

T<sub>4</sub>= Mushroom compost-I @ 5-6 ton /ha

T<sub>5</sub>= Mushroom compost-II @ 5-6 ton /ha

T<sub>6</sub>= Trichocompost @2-2.5 ton/ha

### CHAPTER V SUMMARY AND CONCLUSION

Lentil (*Lens culinaris*) is the most well-known second-largest pulse grain in terms of area and production, but it is also the first widely used low-cost source of plant protein for humans and animals in Bangladesh. Because of its high protein content and quick cooking ability, it is the most important crop. Lentils are a strong source of iron, protein, pantothenic acid, zinc, potassium, and vitamin B6 and have more protein, carbohydrate, and calorie content than other legumes. Fusarium foot and root rot, the most devastating disease that causes huge losses to cultivers, is the greatest yield limiting factor towards lentil production. Several effective pesticides used for the prevention of Fusarium foot and root rot of lentil have been recommended, but they are not regarded long-term solutions because to concerns about cost, exposure to heal the risk, fungicidal residue, and other environmental hazards.

In light of the foregoing, the current study was conducted from October 2019 to May 2020 at the Central Farm And Central MS laboratory of the Department of Plant Pathology, Sher-e-Bangla Agricultural University, Dhaka -1207, to assess the effectiveness of some organic soil amendments in contrasting lentil foot and root rot caused by *Fusarium oxysporum*. The experiment was constructed using a randomized complete block design (RCBD) with three (3) replications. BARI Masur-1 (Utfala) were collected from Bangladesh Agricultural Research Institute (BARI) were sown. Seven treatments were used as soil amendments and they are viz.  $T_0$ = Control,  $T_1$ = Biofumigation with raddish leaf,  $T_2$ = Biochar,  $T_3$ = Vermicompost,  $T_4$ = Mushroom compost-I,  $T_5$ = Mushroom compost-II,  $T_6$ = Trichocompost. All organic soil amendments were applied after final soil preparation and before seed sowing. In this investigation, natural mode of infection was considered on disease incidence. Data were accumulated on disease incidence, plant height, number of branch per plant, number of pods per plant, and yield.

The pathogen was isolated from infected root showing typical foot and root rot stage and identified using cultural and morphological attributes after growing on PDA. The fungal colony in pure culture was first white, cottony, and abundant in aerial mycelium, but as it grew older, it became pink and became entirely dark purple with no aerial mycelium. Conidiophores ranged in length from short to long, branched, and formed a cluster conidiophore known as sporodochia, which bears conidia terminally. Conidia are mostly produced in two forms. Macro conidia are hyaline, enormous multicellular, fusiform, or sickle-shaped produced by sporodochium terminally. Microconidia are round to oval, hyaline, one or two celled, smaller than macro conidia, and generated singly or in chains from aerial mycelium. The fungus was identified as *Fusarium oxysporum* following the keys of Booth (1977; 1971).

Following disease incidence were recorded three times namely 15, 30, 45, and 60 days of their sown. In all counting the effect of various organic soil amendments on foot and root rot disease incidence differed significantly in all counting. The maximum disease incidence was found in control ( $T_0$ ) at 60 DAS which was Trichocompost ( $T_6$ ) had the lowest disease incidence (3.11%) among the organic soil amendments, followed by Vermicompost ( $T_3$ ), Biochar ( $T_2$ ), Biofumigation ( $T_1$ ) and at 60 DAS over control. In terms of growth, yield and yield contributing aspects, the tallest plant (37.33 cm), the maximum no. of branches (28), maximum no. of pods (47.51), the highest seed weight per plant (0.67g), and the yield (564.99 g/plot) were recorded in Trichocompost, ( $T_6$ ) treated plots. The second most tallest plant (30.60 cm), the maximum no. of branches (5.58), maximum no. of pods (43.89), the highest seed weight per plant (0.64 g), and the yield (506.40 g/plot) were recorded in Vermicompost ( $T_3$ ) followed by Biochar ( $T_2$ ) and Biofumigation with radish leaf ( $T_1$ ). Lowest result was recorded in control of all growth, yield and yield contributing parameters.

Trichocompost (T<sub>3</sub>) had the highest yield increase (89.82%). The application of Vermicompost (T<sub>6</sub>) resulted in a yield increase of 70.01%. The seed yield was improved by application of Biochar (T<sub>2</sub>), Biofumigation (T<sub>1</sub>), Mushroom compost -I (T<sub>4</sub>), Mushroom compost -II (T<sub>2</sub>) as organic soil amendment by 67.23, 47.83, 36.72, and 34.98 %, respectively.

Among the treatment organic amendments, few have suppressing effect on several microorganisms. Based on the results of the field experiment, it can be concluded that Trichocompost, Vermicomost, Biochar, or Biofumigation with raddish leaf could be applied on a small scale as an environmentally friendly control of lentil foot and root rot. It was also advised that the study should be conducted over several years in different Agro Ecological Zones (AEZ's) in order to develop a sustainable strategy.

#### **CHAPTER VI**

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## **CHAPTER VII**

## APPENDIECS

# Appendix I: Agro-ecological zones of Bangladesh

ID	Zones/Regions			
1.	Old Himalayan Piedmont Plain			
2.	Active Tista Floodplain			
3.	Tista Meander Floodplain			
4.	Karatoya-Bangali Floodplain			
5.	Lower Atrai Basin			
6.	Lower Punarbhaba Floodplain			
7.	Active Brahmaputra-Jamuna Floodplain			
8.	Young Brahmaputra and Jamuna Floodplain			
9.	Old Brahmaputra Floodplain			
10.	Active Ganges Floodplain			
11.	High Ganges River Floodplain			
12.	Low Ganges River Floodplain			
13.	Ganges Tidal Floodplain			
14.	Gopalganj-Khulna Beels			
15.	Arial Beel			
16.	Middle Meghna River Floodplain			
17.	Lower Meghna River Floodplain			
18.	Young Meghna Estuarine Floodplain			
19.	Old Meghna Estuarine Floodplain			
20.	Eastern Surma-Kushiyara Floodplain			
21.	Sylhet Basin			
22.	Northern and Eastern Piedmont Plain			
23.	Chittagong Coastal Plain			
24.	St Martin's Coral Island			
25.	Level Barind Tract			
26.	High Barind Tract			
27.	North-eastern Barind Tract			
28.	Madhupur Tract			
29.	Northern and Eastern Hills			
30.	Akhaura Terrace			

# Appendix II: 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 (consistency)	: Compact to friable when dry
Soil pH	: 4.47-5.55
Organic matter content	: 0.82

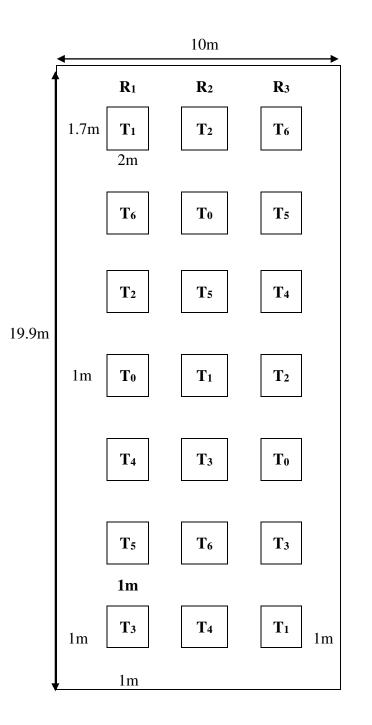
#### Appendix III. Monthly mean weather of the experimental site

Monthly record of maximum and minimum temperature (<sup>0</sup>C), average relative humidity (%), rainfall (mm), average air pressure (mbar) of the experimental site during the period from October 2019 to March 2020 are given below-

Year	Month	Average Temperature (°C)	Average RH (%)	Total Rainfall (mm)	Air Pressure (mbar)
	October	27.7	78	188	1010.1
2019	November	24.9	74	37	1011.5
	December	19.3	74	5	1015.2
	January	18.5	76	21	1014.7
2020	February	21.6	59	1	1014.5
	March	26.4	57	30	1010.7

Source: Bangladesh Meteorological Department (Climate division), Agargaon, Dhaka-1207.





Total Area-19.9m × 10m = 199 m<sup>2</sup>

Plot Size  $-2m \times 1.7m$ 

Component	Amount	
Potato extract	200 g	
Dextrose	20 g	
Agar powder	20 g	
Distilled Water	1 L	

## Appendix V. Composition of culture media (PDA)

## Appendix VI. Composition of lectophenol cotton blue (LCB)

Component	Amount	
Cotton blue	0.05g	
Phenol crystals	20g	
Lactic acid (S.G. 1.21)	20g	
Glycerol	40g	
Distilled water	20ml	

Parameters	LSD (0.05)
Disease incidence at 15 DAS	1.7199
Disease incidence at 30 DAS	2.2977
Disease incidence at 45 DAS	1.7977
Disease incidence at 60 DAS	1.39
Plant height at 45 DAS	1.62
Plant height at 60 DAS	2.1058
Plant height at 75 DAS	2.4068
No. of branch per plant at 45DAS	0.3133
No. of branch per plant at 60 DAS	0.4414
No. of branch per plant at 75DAS	0.6922
No. of pods per plant	3.7076
Yield (g/plant)	0.694
Yield (g/plot)	36.608

Appendix VII. LSD value for different parameters at 5% level of significance

Appendix VIII. Mean square value and degree of freedom (DF) of disease incidence of foot and root rot disease in lentil at different DAS from Analysis of variance (ANOVA)

Source of variation	DF	Mean square value of disease incidence			
		15 DAS	<b>30 DAS</b>	45 DAS	60 DAS
Treatment	6	258.025	442.692	599.363	866.613
Error	12	0.935	1.821	1.021	0.611

Appendix IX. Mean square value and degree of freedom (DF) of plant height of foot and root rot disease in lentil at different DAS from Analysis of variance (ANOVA)

Source of variation	DF	Mean square value of plant height		
		45 DAS	60 DAS	75 DAS
Treatment	6	47.129	70.794	124.228
Error	12	0.836	1.401	1.830

Appendix X. Mean square value and degree of freedom (DF) of no. of branches per plant of foot and root rot disease in lentil at different DAS from Analysis of variance (ANOVA)

Source of	DF	Mean square value of no. of branches/ plant			
variation					
		45 DAS	60 DAS	75 DAS	
Treatment	6	2.302	5.119	5.507	
Error	12	0.031	0.061	0.151	

Appendix XI. Mean square value and degree of freedom (DF) of yield parameters of foot and root rot disease in lentil from Analysis of variance (ANOVA)

Source of variation	DF	Mean square value of yield parameters			
		No. of Yield/plant Yield/ Plot			
		pods/plant			
Treatment	6	314.786	0.090	23006.3	
Error	12	4.343	0.001	423.4	

# Appendix XII. Seed sowing in the field



Appendix XIII. Experimental field view



Appendix XIV. Data collection in the field

