MANAGEMENT OF FUSARIUM WILT OF BRINJAL (*Fusarium oxysporum* f. sp. *melongenae*) BY USING DIFFERENT ORGANIC AMENDMENTS

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CERTIFICATE

This is to certify that the thesis entitled, "**MANAGEMENT OF FUSARIUM WILT OF BRINJAL (*Fusarium oxysporum* f. sp. *melongenae*) BY USING DIFFERENT ORGANIC AMENDMENTS**" 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 (MS)** in **PLANT PATHOLOGY**, embodies the result of a piece of bona-fide research work carried out by **JANNATUL MAUYA**, Registration no. **20-11093** 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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DEDICATED TO MY BELOVED FAMILY AND HUSBAND

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MANAGEMENT OF FUSARIUM WILT OF BRINJAL (*Fusarium oxysporum* f. sp. *melongenae*) BY USING DIFFERENT ORGANIC AMENDMENTS

ABSTRACT

A field experiment was conducted in the central farm of Sher-e-Bangla Agricultural University and lab experiment was conducted in the lab of Department of Plant Pathology, Sher-e-Bangla Agricultural University, Dhaka during the period from September 2021 to April, 2022. The study was carried out to evaluate the effect of selected treatments viz. T_0 = Control ; T_1 = Spent Mushroom Substrate (SMS) ; T_2 = Vermicompost ; T_3 = Poultry manure; T_4 = Biochar ; T_5 = Spent Mushroom Substrate + Biochar ; T_6 = Spent Mushroom Substrate + Poultry manure ; T_7= Spent Mushroom Substrate + Vermicompost for the management of Fusarium wilt of brinjal. In response to different selected treatments, yield and yield attributes were recorded. On the basis of visible symptoms infected plants and percent disease incidence were also recorded. The lowest disease incidence was found in T_5 (Spent Mushroom Substrate + Biochar) followed by T_6 (Spent Mushroom Substrate + Poultry manure) and the highest disease incidence was found in T_0. The effect of different treatments on yield and yield contributing characters also studied and significance variation was observed. For yield contributing characters the lowest result was found in untreated control T_0 and the highest result was found in T_5 which was statistically similar with T_6 .In response to different selected treatments for yield, the highest performances was observed where Spent Mushroom Substrate + Biochar was used. From the findings of the present study, it may be concluded that Spent Mushroom Substrate + Biochar and Spent Mushroom Substrate + Poultry manure can be used as ecofriendly approach for the management of fusarium wilt of brinjal. However, further investigation is needed to justify the present findings.

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INTRODUCTION

Brinjal (*Solanum melongena* L.) is the most popular, widely available and reasonably priced vegetable that belongs to the family solanaceae. Brinjal is one of the most significant vegetables grown worldwide including Bangladesh. Depending on the location (i.e., differences between the English speaking countries) and type of fruit, the Eggplant usually known as Brinjal in south Asia (especially Pakistan, India, and Bangladesh), and called aubergine in Europe, melongenae in West Indies, Guinea squash in America and patlican in Turkey. In Bangladesh, "Begoon" (also known as brinjal or eggplant) is a common and beloved vegetable that has a connection to the social, cultural, and rural residents' financial circumstances.

Since ancient times, brinjal has been a common vegetable in our diet. It is one of the utmost vegetables and its production ranks third among all vegetables in the world. Although it is grown all throughout the world, Asia has the highest concentration. In a subtropical country like Bangladesh, eggplant is grown all over the country on medium high land to high land in both Rabi and Kharif seasons. In the Kharif (summer) season in Bangladesh, 49406.92 acres were cultivated, with total production at 209541.17 MT and in the Rabi (winter) season 84536.65 acres were cultivated, with total production 409001.46 MT (BBS, 2022).

The fruit of the brinjal is very low in calories and has a healthy amount of minerals. Brinjal contains all of the essential vitamins, minerals, and nutrients, including iron, calcium, potassium, magnesium, nutritious fiber, protein, and antioxidants (Apendix-4). It also contains certain phytochemicals with scavenging properties. Brinjals are rich in anthocyanin chemicals. The strong protective effects of anthocyanin against diabetes, cancer, cardio vascular disease. (Naeem and Ugur, 2019).

However, production and yield rate of brinjal in Bangladesh is appalling compared to other countries like India, Japan, China due to spread of various diseases. Despite favorable environmental conditions, theper hectare yield of brinjal is far lower than its potential production for a number of reasons. Diseases are one of the main factors for low production.

Diseases caused by fungi, bacteria, viruses, and nematodes severely reduce the amount of brinjal produced (Zeeshan *et al*., 2016). Wilt of brinjal is one of the most harmful disease among all the diseases of brinjal that causes great economic loss of brinjal production. Fusarium wilt initiated by *Fusarium oxysporum* f. sp. *melongene* are the most devastating one. Both the quality

and yield of the brinjal production are decreased by *Fusarium oxysporum* f. sp. *melongenae*. This pathogen was initially reported in Japan and next in China (Rao, 2022). The severity of fusarium wilt in brinjal is 10%-90% (Sahoo, 2022).

Bangladesh ranks quite poorly when compared to other nations in terms of production and yield rate due to a lack of adequate expertise. Fusarium wilting generally causes 20–30% of eggplant plants to die and it may turn into epidemic during November to December (Adhikary *et al*., 2017). Since the pathogen is soil-borne and has a large host range, comprising several hundred species from 44 families of plants, it is challenging to control. Infection is occurred through Root-to-root transmission, soil movement and dispersal by agricultural equipment, and insect transmission (Rao *et al*., 2019).

This pathogen attacks all phases of growth (vegetative and regenerative) and survives on plant debris for long periods. The signs include leaf chlorosis, slowed development, vascular vessel discoloration, and wilting, which leads to plant death (Soleha *et al*., 2022).

Fusarium oxysporum can survive in the soil for a long period of time without any host. Because *Fusarium oxysporum* is a soil-borne pathogen that makes it challenging to regulate. Several control measure such as crop rotation, the use of various resistant cultivars, soil sterilization and solarization and the application of fungicides are some useful or effective management strategies for Fusarium wilt. However, due to the possible negative impacts of fungicides on the environment and human health as well as their unfavorable effects on nontarget organisms, the extensive use of chemical fungicides has been a source of public worry and security (Basco *et al*., 2017).

Biological control could be successful alternative to chemicals. Additionally, compared to conventional pesticides, biological pest control is less hazardous to the environment. On the other hand, some biocontrol agents (BCAs) indirectly promote plant growth while the pathogen is present, reducing the pathogen's negative impacts via mechanisms such nutrient competition, ecological site competition, induced systemic resistance, and antibiosis (Lugtenberg *et al*., 2009). Recent studies have found that biological control agents
(BCAs) for plant diseases are an effective substitute for synthetic pesticides because of their high level of biosafety and minimal negative effects on the environment. Several biocontrol agents such as spent mushroom substrate, poultry refuse, vermicompost have the ability to manage some soil borne pathogen.

When SMS is allowed to accumulate as garbage in the environment, it can act as a source of pollution, endangering the environment. Yet, this negative effect can be advantageous when utilized as a substrate for cultivating agricultural crops. After the harvest of the mushrooms, spent mushroom substrate (SMS) is a waste byproduct of the mushroom industry. When different flushes of mushrooms have been harvested, spent mushroom compost (SMC), also known as spent mushroom substrate (SMS), is the residue of trash. (Jonathan *et al*., 2011). 1 kg of mushroom production will generate about 5kg of spent mushroom substrate (SMS) (Zied *et al*., 2020). Because of its high cation exchange capacity (CEC), slow mineralization rate, and rich nutritional status, the SMC has been discovered to be an excellent source of nutrients for agriculture (Verma *et al*., 2020). It naturally controls soil infections, reduce damage of plants and reduce crop production.

SMS is rich in diverse microorganisms, such as disease antagonistic bacteria and fungus. It is biodegradable, safe to apply and less expensive to develop (Adedeji *et al*. 2016). Utilization of Spent Mushroom Substrate is ecofriendly approach to manage different soil borne disease. Its use in organic agriculture, zero waste integrated farming systems, as well as integrated disease management is receiving attention recently.

Vermicompost is one of the greatest organic manures for enhancing crop yield. Vermicompost has been discovered to be a tool for reducing the dangers of chemical fertilizers. Vermicomposting is the natural process of rotting or decomposition of organic matter by the activity of earthworm under controlled conditions (Gudeta *et al*., 2022)

The organic matter resources found in vermicompost are abundant and have the unique potential to enhance the chemical, physical, and biological properties of soils or growing media. Vermicompost's organic carbon slows the release of nutrients into the system so that they can be absorbed by the plant. (Gandhi *et al*., 2012). Vermicompost is also useful as it increases soil porosity, aeration and water holding capacity. Vermicompost enriches the soil by adding additional nutrients that chemical fertilizers do not contain. They may be very suppressive against soil borne diseases when employed as soil additives. (Mamta *et al*., 2012 and Geroche *et al*., 2019) Vermicompost contains a large number of antagonists. Actinobacteria, Chloroflexi, Saccharibacteria, and Planctomycetes grew more readily under the vermicompost treatment than under the control treatment and reduce the growth of harmful pathogen. Vermicompost treated area showing disease incidence of *F. oxysporum* 36.5% - 73.9% lower than the control treatment (Wang *et al.,* 2021)

Poultry manure is an effective soil amendment that supplies nutrients for growing crops. Poultry manure is the term for the waste products produced by chickens and other domesticated birds raised for their eggs, meat, or feathers. Poultry manure has a high nutritional value since it contains nitrogen, phosphorus, and potassium. (Richa *et al*., 2020). Poultry manure act as a soil conditioner to reduce the bulk density of the soil. Poultry manure also has the ability to reduce the activity of soil bone pathogen. The incidence of wilt (*F. oxysporum*) can be reduced 41.15% by poultry manure (Jat *et al.,* 2017).

Biochar is a type of charcoal that is produced through a process called pyrolysis, which involves heating organic materials (such as wood chips, agricultural waste, or animal manure) in the absence of oxygen. Biochar has been utilized to boost agricultural productivity, enhance soil health, and lower greenhouse gas emissions. (Akanmu *et al*., 2020). Biochar enhence plant defenses against many soil- and airborne diseases. In comparison to a control, adding biochar to asparagus field soil at 1.5 and 3.0% (wt/wt) led to proportionate increases in root weights and linear decreases in the percentage of root lesions brought on by *Fusarium oxysporum* f. sp. *asparagi* and *F. proliferatum* (Elmer *et al*., 2011). It was noted that the growth rate of *Fusarium* spp. in soils treated with biochar was 50% lower than in soils not treated with biochar.

This study proposes the use of spent mushroom substrate to combat the pathogen *Fusarium oxysporum*. This research work presents the effectiveness of Spent Mushroom Substrate and other soil amendments for the management of wilt of brinjal (*Fusarium oxysporum*).

Objectives:

The research work was carried out to achieve the following specific objectives:

- To estimate the disease incidence of fusarium wilt of brinjal in field at natural condition; and
- To evaluate the potentiality of selected organic amendments to control Fusarium wilt disease of brinjal.

REVIEW OF LITERATURE

Brinjal (*Solanum melongena* L.) is a highly well-known and widely grown vegetable crop because of its flavor, nutrition, and therapeutic value. There are a number of diseases that affect brinjal, the most severe of which is brinjal wilt. Fusarium Wilt of brinjal caused by *Fusarium oxysporum f.sp. melongenae* is a soil borne disease that significantly hinders the production of brinjal. Even after the rotation of non-susceptible crops, the disease's pathogen continues to exist for a considerable amount of time as chlamydospores in plant and soil debris. So it is quite difficult to handle this disease. Some available and important findings on various aspects of management of fusarium wilt have been compiled and presented below.

2.1 Importance of brinjal

Brinjal, often known as eggplant (*Solanum melongena* L.), is a popularly produced vegetable that is a member of the Solanaceae family. Generally the crop grown in Asian nations; most likely a native of South Asia. It is grown in all of India on an area of 668.72 thousand ha, producing 123.99 thousand tonnes per year with a yield of 18.53 million tonnes per hectare. In 2016–17, the area, production, and productivity of brinjal in Maharashtra were 221,410 ha, 43328000 tonnes, and 19,68M tons ha^{-1} respectively. (Rao *et al*., 2022)

Brinjal, a well-liked vegetable, is a significant source of income for Bangladeshi small-scale farmers who lack resources. According to a survey of Bt and non-Bt farmers, Bt brinjal varieties produced an average yield and revenue that was 21.7% greater than non-Bt types. Using Bt brinjal increased revenue by 1.7% per tonne, reflecting varying levels of consumer and trade buyer acceptance. Several consumers were willing to pay more for Bt brinjal than non-Bt brinjal since the fruit was less damaged (Shelton *et al*., 2020).

Generally eggplant is warm season plant. It often grown two or three times per year. So, it is available in markets all throughout the year. Egg to long club-shaped, eggplants can be found in a range of colors and shapes, with colors ranging from white, green, and yellowish to various shades of purple pigment, practically black. The fruit serves both culinary and medical purposes.The fruit of the eggplant has very few calories and a healthy mineral composition. Magnesium, manganese, potassium, and copper are abundant in eggplants and are vital nutrients for strong bones. As an additional Fe chelator, eggplant is recommended in particular for female teenagers, breastfeeding moms, and pregnant women. The Fe found in eggplant has the capacity

to treat prenatal anemia, amenorrhea. The fruits are useful in the treatment of a variety of ailments such as asthma, dysuria, dysentery, high blood pressure and also to cure osteoporosis, arthritis, diabetes and bronchitis,heart ailments and stroke (Naeem *et al*., 2020).

In Bangladesh as well as the rest of the world, brinjal (*Solanum melongena* L.) is a significant vegetable due to its commercial and nutritional importance. Brinjal is by far the most important vegetable, accounting for 41% of the weight of all produced vegetables and taking up 19% of the land needed for its cultivation. Because to increased profits, a relatively quick growth rate, little risk, and early technological adoption, Farmers are egarly cultivated brinjal over the years. Brinjal consists of about 92.7 percent of water and is superior in terms of fiber, folic acid, manganese, thiamin, vitamin B6, magnesium and potassium levels to that of most other vegetables. It offers 25 calories per serving and no fat (Rahman *et al*., 2016).

Brinjal is a significant and well-liked vegetable in Bangladesh that is eaten all year round. Based on the time of production, brinjals are divided into two groups. These are brinjal from the Rabi and Kharif seasons. Although it is generally accessible throughout the year, its supply is at its highest from December to April. Minerals and vitamins are abundant in brinjal. 100 g of edible brinjal has 24 kcal of food calories, 1.4 g of protein, and 18 mg of calcium.. In addition, compared to most other vegetables, brinjal is superior in terms of fiber, folic acid, manganese, thiamin, vitamin B6, magnesium, and potassium. It also contains nearly 92.7 percent water (Hasan and Bai, 2016).

Solanum melongena L., a member of the Solanaceae family, is a common and well-liked warm weather vegetable crop farmed extensively in India. It is a significant source of income for marginal and small farmers. Unripe fruit is utilized in cooking for a variety of regional specialties. Many plant parts are used to treat bronchitis, diabetes, cholera, dysuria, dysentery, toothaches, and skin infections. The plant also has important therapeutic potential (Sain and Pandey, 2016).

Brinjal is commonly known as poor man’s meat because of its inherent high nutritive value, vitamin content and dietary fibers. Moreover, brinjal has some medicinal value. It can block free radicals, can help to control cholesterol levels and brinjal is also a source of folic acid and potassium. (Barman *et al.,* 2013)

2.2 Importance of fungal wilt disease (*Fusarium* spp.)

Akter *et al*., (2021) described that eggplant cultivation is significantly impaired by wilt complex caused by *Fusarium oxysporum*, *Ralstonia solanacearum* and *Meloidogyne sp*. The total yield of brinjal production is reduced 20% - 30% due to the destructive disease called fungal wilt. It may turn into epidemic in the winter season, even complete crop failure may occure due to the epidemic condition of this disease.

Das (2021) stated that there are more than 20 distinct diseases that affect brinjal. Fusarium wilt is the most devastating worldwide. Members of the Fusarium genus are common soilborne pathogens that affect a variety of food and horticultural crops and produce deadly vascular wilts, rots, and damping off diseases. A significant, common fungus that lives in soil and is noted for its phylogenetic variety is *Fusarium oxysporum*.

Adhikary *et al*., (2017) highlighted the management of Fusarium wilt as the most detrimental interruption to better eggplant production. Farmers typically bear the brunt of this deadly disease. Fusarium wilt of brinjal caused by *Fusarium oxysporum* f. sp. *melongenae*. Farmers regularly report that Fusarium wilting causes a 20–30% reduction in brinjal production.

Basco *et al*., (2017) observed that Fusarium wilt is one of the most serious diseases of tomato that affects its yield. This disease is caused *by Fusarium oxysporum* and the yield loss due to this disease is 25.14- 47.94 % in Uttar Pradesh. *Fusarium* spp. are well established soil borne pathogens in all soil type throughout the world. *Fusarium* spp. are saprophytes and are able to grow on soil organic matter for a prolonged period.

Islam *et al*., (2017) described among the many obstacles to the cultivation of brinjal, diseases play an important role. Over 70 different diseases are known to affect brinjal. Fusarium wilt of eggplant has been considered one of the main obstacles to eggplant cultivation in Bangladesh among those ailments.

Faruq *et al*., (2014) described that *Fusarium oxysporum* f. sp. *melongenae* is a soil-dwelling fungus that causes Fusarium wilt of eggplant, which is very prevalent in eggplant-growing regions and can result in significant yield loss.

Jatav *et al*., (2013) stated that in eastern Rajasthan and Uttar Pradesh, the wilt disease of brinjal is highly widespread and results in significant losses. The severity of disease is greater in soils with pH values below 6.4 and over 7pH. Crop loss was between 5% and 60%. In Rajasthan,

where disease infection rates varied from 70 to 80% in vegetable fields, Mathur and Prasad (1964) recorded an average loss of 20%.

Barman *et al*., (2013) stated that over the growing seasons, brinjals are susceptible to diseases and disorders. The brinjal wilt pathogen *Fusarium oxysporum* f. sp. *melongenae* is quite damaging. This fungus inflicts a disease that is characterized by wilted plants, yellowed leaves, and little or no crop production.

2.3 Symptoms of *Fusarium* wilt

Eggplant (*Solanum melongena* L.) is a very popular vegetable in our country. But the production of eggplant is hampered because it suffers from various disease. Despite favorable environmental conditions, the yield of brinjal per hectare falls significantly below of its maximum production for a variety of reasons. One of the main causes of low output is disease. Diseases caused by fungi, bacteria, viruses, and nematodes severely reduce the amount of brinjal produced. Wilt is one of the most destructive disease that affects brinjal production and costs producers a lot of money. The production of brinjal is severely hampered by the significant disease called fungal wilt (*Fusarium oxysporum* f. sp. *melongenae*), which is a soil-borne disease and difficult to manage.

Chaterjee, *et al*., (2021) described that wilting symptoms manifest as isolated areas with fewer or more circular outlines that increase as the condition worsens. Starting from the lowest leaflet surface, curling moves upward. Hence, the crown may begin to bend and plants may die. Plants get discolored or brown because their root systems are unable to grow normally. Roots may become completely or partially discolored.

Khan *et al*., (2019) stated that Fusarium wilt is characterized by yellowing of the leaves and drooping of the apical shoot, which ultimately results in the death of the entire plant. The most noticeable symptom of Fusarium wilt is epinasty of leaves and partial stunting of eggplant. It first manifested as minute vein clearing on the outer regions of immature leaves, which was followed by older leaves bending downward. At the seedling stage, plants exhibit *Fusarium oxysporum* infections, which may cause wilting and plant death. The bottom surface of mature leaves turns yellow, adventitious roots begin to emerge, leaflets or newly formed stems may wilt, and there may also be defoliation, browning, or necrosis at the leaf margins. As a result, the entire plant may die.

Naziya and Sharada, (2018) described that a destructive disease of brinjal is Fusarium wilt that caused by *Fusarium oxysporum* f.sp. *melongenae*. It is a soil-borne fungus that invades through vascular bundles and blocks the xylem transport system, causing severe wilting and death of the plant's upper sections.

Biswas and Ghosh, (2018) reported that the fungus that causes fusarium wilt of eggplant, *Fusarium oxysporum* f. sp. *melongenae*, is the most devastating. By obstructing the xylem transport system, the soil-borne fungus invades the vascular bundles, severely wilts, and eventually kills the above-ground part of plants.

Adhikary *et al.,* (2017) described that in November and December, the crop may completely collapse due to an epidemic of the disease. The fungus clogs the plant's vascular system, causing it to die from a lack of water and nutrients. The fungus primarily attacks the plants during the period of growing up.

Kareem and Al-Araji, (2017) stated that The infected plants were diagnosed based on the symptoms that appeared on the vegetative and root, which included yellowing, drooping and falling of leaves, death of some branches, reddish-brown streaks were visible in the vascular tissues when cut with a knife and finally death of plants.

M. Patil *et al*., (2017) described that spores of this fungus live in the soil for many years, the disease clogs the xylem transport system and causes severe wilting and death of brinjal plants. It is difficult to manage using typical chemical fungicides.

Abdel-Monaim *et al*., (2014) described that Fusarium wilt is a disease that affects eggplant yield. The most dangerous pathogen of Fusarium wilt of eggplant is *Fusarium oxysporum* f. sp. *melongenae*. By obstructing the xylem transport system, the soil-borne fungus invades the vascular bundles, causing severe wilting and death of the plant's aboveground portions.

Jatav *et al*., (2013) describe that the disease's initial signs include clearing of the veinlets, although the major veins remain green. This is followed by unilateral yellowing of the younger leaves, followed by withering and death, which starts in the older leaves and spreads up to the main stem and eventually the entire plant. Brown staining is visible in the xylem channels of diseased plants. The root system has been greatly decreased, and their color has also changed to a light black. The root expands and becomes spongy.

2.4 Pathogenic description of *Fusarium oxysporum*

Fungal wilt of brinjal is a major disease that affect the brinjal production globally. The disease is difficult to manage because the pathogens can persist in the soil for several years by producing chlamydospores without any host. Therefore, alternative approaches are needed for effective management of this disease.

Zheng *et al*., (2022) stated that *Fusarium* spp. create a carcinogenic mycotoxin, which directly threatens food security and human health. Furthermore, *Fusarium* phytotoxicity is thought to be a factor in the severity and progression of plant diseases. They also observe microconidia were likewise single-celled, ellipsoidal, 0-1 septate, and 5–14 × 2.5–4.5 µm. Macroconidia were falcate, curved to the ventral side, with papillate apical cells and foot-shaped basal cells, 1-4 septate (usually 3 septate), hyaline, smooth, and thin-walled, 21–42 × 3–6 µm. Chlamydospores were globose to subglobose, with an average diameter of 10 µm and hyphae that branched at acute angles.

Soleha *et al*., (2022) stated *Fusarium oxysporum* is a soil-borne pathogen with a diverse host range that is ubiquitous in many areas around the globe, including forests and industrial plantations. This pathogen targets all stages of growth (vegetative and regenerative) and can live for long periods on plant waste. In plants, it produces vascular wilt or root rot disease. The symptoms include leaf chlorosis, slowed development, staining of the plant's vascular vessels and withering, which leads to death. This species is a hazardous pathogen for plants grown in both open fields and greenhouses.

Chaterjee *et al*., (2021) stated about the fungus that chokes the plant's xylem-phloem, causing death of the plant from lack of water and nutrients. The pathogen is primarily soil-borne and produces macroconidia, microconidia, and chlamydospores, so controlling this destructive disease is extremely challenging. These viable and thick-walled chlamydospores containing lipid-like substances survive in the soil for a long time.

Himabindu *et al.,* (2021) described that there are over 100 Fusarium vascular wilts in the world. This disease has sparked global interest because it has been documented in 32 nations, with the disease being most severe in countries near the equator. *Fusarium oxysporum* or *Fusarium solani* produce Fusarium wilt. There are more than 100 Fusarium vascular wilts found worldwide. This is of global significance, with approximately 32 countries reporting on this ailment, which is most severe in areas near the equator. The pathogen is widely adaptable to

climate conditions and has a polyphagous character. Single method is insufficient to manage this disease.

Hassan (2020) described that *Fusarium oxysporum* is a damaging soil-borne fungal pathogen that causes tomato wilt disease. During age, it generates white aerial mycelium with pink, orange, red, blue, and purple coloring. Depending on the culture circumstances, *Fusarium oxysporum lycopersici* can produce three forms of asexual spores: macroconidia, microconidia, and chlamydospores. Macroconidia form long, sickle-shaped, thin-walled spores with several septa. Microconidia are single-celled, oval-shaped spores that are numerous. A unique feature is the formation of microconidia on short monophialides. Chlamydospores are spherical and thick-walled; dormant spores form singly or in pairs. On old culture, it is normally generated in 2-4 weeks.

Verma *et al.,* (2017) described about the nature of *Fusarium* spp. and stated that the fungus is a slow growing species. The *Fusarium* spp. is typically described as light pink, creamish white to creamy, and light purple to violet in colour. The fungus produced macro and micro conidia which included thin-walled, 3-5 septate, fusoid falcate macro conidia with a somewhat hooked apex and pedicillate base.

Salim and Simon (2015) described *Fusarium oxysporum* is a global fungus with numerous harmful forms. It persists in soil as chlamydospores and mycelia. The mycellium is septate, hyaline, branching, and intracellular. It creates problems at all stages of plant growth, from nursery through flowering. Clearing of veinlets and drooping of petioles of immature plants cause yellowing of lower leaves, as a result plant wilts and dies prematurely. A cross section of the lower stem reveals browning of the vascular system. In moist weather, pinkish mycelial layers of fungal mycelial development can be visible on dead plants.

Altinok (2005) stated that the fungus *Fusarium oxysporum* was identified by the formation of typical three to five septate, sickle-shaped macroconidia with a foot-shaped basal cell, ellipsoid microconidia borne in false heads on short monophialides, and chlamydospores in culture and on PDA media the fungus formed a typical cream-colored colony with purple pigmentation on the reverse side.

Miller *et al*., (2005) stated that *Fusarium oxysporum* that causes Fusarium wilt in solanaceous crops. *F. oxysporum* f. sp. *lycopersici* (tomato), *F. oxysporum* f. sp. *melongenae* (eggplant), and *F. oxysporum* f. sp. *vasinfectum* (pepper) are the different strains of *F. oxysorum*. All

Fusarium wilt diseases are soilborne and often unique to their hosts. The fungus survives in the soil as chlamydospores and enters the host roots either directly or through wounds.1

2.5. Study of pathogen isolation

Ram *et al*. (2022) isolated the fungus from fresh diseased solanaceous plants that were collected from the field of different location of Bundelkhand location. Paper bags were used to collect samples of the root tissues that had been affected by wilt in order to isolate *Fusarium oxysporum* f. sp. *ciceri.* They followed tissue isolation approach after through surface sterilization of root sections (2-3 mm size) using 0.1 percent mercuric chloride solution for a minute. To eliminate the mercuric chloride treated parts were then thoroughly rinsed three times in sterile distilled water. Then put into petri dishes that contained PDA media that had solidified. The inoculation plates were then incubated at 25±2° temperature and checked regularly. They used single hyphal tip cut techniques for purification of the fungus. Based on the shape of the spores and conidiophores, *Fusarium oxysporum* f. sp. *ciceri* was identified.

Muhammad *et al*., (2022) isolated the fungus *F. oxysporum* from infected chilli plants exhibiting wilt symptoms. The diseased sample was divided into small pieces measuring 4-6 mm, cleaned with distilled water, surface sterilized for 30 seconds with 1% $NaOCl_2$, and then cleaned three more times with sterilized distilled water before being set aside to dry. With the aid of sterile forceps, the samples were deposited on PDA media, incubated at 28 ± 2°C and monitored every day for colony growth. The fungus was also subcultured in order to purify the culture. Under a light microscope, *F. oxysporum* was identified based on the pathogen's morphological characteristics, including as colony growth, color (purple and white), conidiophores, and microconidia.

Das, (2021) collected wilted brinjal plants from farmers' fields and for isolation of the fungus brought to the pathological laboratory of Botany Department, Gauhati University, Guwahati, Assam. To get rid of the dust and surface contaminants, the diseased parts were surface sterilized with 0.01% mercuric chloride ($HgCl_2$) solution for 60 seconds and rinsed 4-5 times in sterilized distilled water. By placing the pieces on sterilized filter paper, more moisture was extracted from the bits. The fragments were then placed in sterile petri plates with Potato Dextrose Agar (PDA), where they were cultured for a period of time at room temperature (28°C). On PDA 28°C, the cultures were purified using the hyphal tip technique. He used single spore method to create the pure culture of the isolated fungus *F. oxysporum*.

Siddique *et al.,* (2019) isolated *F. oxysporum* f. sp. *lycopersici* from tomato plants that were naturally infected and were gathered from two distinct fields, namely the central and horticulture fields. Little sections of the plant parts with brown vascular tissue discolouration were removed (1 cm). These pieces were evenly spaced and aseptically put to sterile potato dextrose agar in each petri dish. The inoculation plates were then incubated at 28 °C. Based on the characteristics listed by Booth, the culture, *F. oxysporum f.sp lycopersici*, was identified.

Biswas and Ghosh (2018) isolated the pathogen from diseased plants by using a normal tissue culture procedure. The related pathogen was then identified using cultural, morphological, and microscopic characteristics.

Basco *et al*., (2017) isolated the fungus from infectious tomato stem and root tissues exhibiting indications of wilt, that were separately collected from an agricultural area at Banaras Hindu University in Varanasi, India. Surface sterilization with 3% sodium hypochlorite was performed on tissue bits for 3 minutes, followed by three washes with sterile distilled water. They were then individually placed on potato dextrose agar (PDA) media and cultured in a BOD incubator for 5 days at a temperature of 25 °C. By transferring the tip of the mycelia into new PDA plates and maintaining the pathogen on PDA slants that were preserved at 4°C as stock cultures for further research, the pathogen was individually purified. Based on physical traits such as micro-conidia and macro-conidia, the culture was identified.

Barman *et al.,* (2013) isolated *Fusarium oxysporum* from diseased brinjal plants. Sodium hypochlorite (1%) was used to disinfect a piece of an affected part for five minutes. The fragment was placed on modified peptone pentachloronitrobenzene agar (PPA), a medium designed specifically for *F. oxysporum*. Five days of incubation at 28 ºC resulted in the development of *F. oxysporum* growth surrounding the component. To validate the growth of *F. oxysporum* mycelium, a portion of the established mycelium from the margin was subcultured three times on potato dextrose agar (PDA) by incubating the plates at 25 ºC for seven days. A portion of the isolated *F. oxysporum* mycelium was suspended in a flask containing 250 ml of potato dextrose broth and was incubated until the liquid medium was brown-pink in colour.

Nirmaladevi and Srinivas (2012) isolated *F. oxysporum* from a wilted tomato plant. Rinsed root and stem tissues under running water to facilitate isolation. The plant fragments removed from the upper and lower taproots were surface sterilized for 1 to 2 minutes in a 1% NaOCl solution, rinsed twice in sterile distilled water, and dried between sterile filter papers. On potato dextrose agar, pieces of tissues that had been surface infected were put (PDA). The plates were incubated

for 7–10 days at 28 ± 2 °C. They used dilution plate technique for soil samples from the rhizosphere. Then incubated plated soil dilutions for five days at 28 ± 2°C.

2.6. Pathogenicity study of *F. oxysporum*.

Zheng *et al*., (2022) tested the pathogenicity on eight *F. oxysporum* isolates in triplicate. During an in-vivo experiment, healthy *P. sinese* seedlings that were 6 weeks old were poked with sterile needles and had their stems soaked in a spore suspension for 30 minutes. As a negative control, distilled water was used. All plants were grown on sterilized soil in a greenhouse at 24°C with a 12h/12h light/dark cycle. The petri plate used for the in vitro test had previously been cleaned with 75% alcohol and cushioned with autoclaved filter paper that had been kept moist with sterilized distilled water. *P. sinese* leaves were separated, poked, and inoculated as previously mentioned. In the biochemical incubator, culture dishes had been set up. The illumination condition was set to 24 hours of darkness, and the temperature parameter was changed to 25 °C. In order to confirm Koch's hypotheses, fungal pathogens were once again isolated from the symptomatic tissues of the infected plants and identified using cultural traits and DNA analysis.

Soleha *et al.,* (2022) conducted the experiment on *A. mangium* seedlings 30 days after sowing that had been sown in plastic containers with 200 g of sterilized peat soil. By putting 5×5 mm agar pieces of the fungal colony in the potato dextrose broth (PDB) medium, fungal isolates were produced. Large numbers of conidia were produced throughout the three-day incubation period utilizing a shaker at a speed of 120 rpm. While the uninoculated control was sterile distilled water, this suspension was utilized as an inoculum by dispensing 1 ×106 cfu g^{-1} (colony forming unit/g) onto a soil media. Ten test plants' soil was inoculated with each isolate, and the procedure was repeated once. The quantity of disease incidence was measured. For each seedling the disease severity was calculated using a score of 0−4, where 0 = no disease/healthy seedling, 1 = yellow leaves, 2 = yellow leaves and slightly wilted, 3 = severe wilt and 4 = dead seedling. The observation of the plant was done over 30 days after inoculation.

Das (2021) used Root-dip method for the pathogenicity test of the fungus *F. oxysoporum*. Healthy brinjal seedlings were uprooted from sterilized soil when they had six leaves and the damaged roots were immersed in a conidial suspension (106 conidia/ml) for 10 minutes while the control plants were plunged in sterile tap water. After that, seedlings were placed into clean pots. After three weeks of inoculation, symptoms were seen on the plants, and the pathogens were re-isolated.

Kalman *et al.,* (2020) conducted an onion bulb pathogenicity experiment on Riverside (Orlando) cv. and Noam cv. (white and red onion cultivars, respectively) for the Koch's postulates accomplishment. Isolates were used six times to complete the full test in a sterile biological hood. A stock of 106 spores/ml (in sterile water) was made from each isolate using a five day old colony that had previously been cultivated on a PDA at 28 ± 1 °C in the dark. The bulbs were cleaned in 70% ethanol after the outer scales were removed, and then they were dried. The basal plate was stabbed five times at a depth of 5 mm using a sterile pipette tip (10 mm in diameter) to do the injection. To maintain a moist environment and avoid unintended contamination, each bulb was maintained separately in a closed sterile plastic bag and housed in a dark, temperature controlled incubator at 28°C. After two weeks of incubation, the fungus from each infected onion was re-isolated on PDA and identified to support Koch's postulate.

Kareem and Al-Araji (2017) investigated the virulence of *F. oxysporum* and the results showed that all isolates of the fungus were harmful to eggplant plants, but to various degrees. The four isolates of *F. oxysporum* with the highest virulence were F5, F6, F13, and F14, infecting eggplant plants at two different times: before they emerged (83.3%, 83.3%, 86.7%, and 83.3%) and after they emerged (90.0%, 90.0%, 83.3%, and 76.7%). Root recognition, root surface attachment and colonization, root cortex penetration and colonization, and occasionally hyphal growth within the xylem vessels were the stages of the *F. oxysporum* infection process. Then, during root penetration and colonization, the fungus secretes a number of cell wall-degrading enzymes, including polygalacturonases, pectate lyases, xylanases, proteases, and cellulase.They reported that the pathogenic fungus *F. oxysporum* was highly virulence on eggplant and other solanaceae family.

Abdel-Monaim (2014) observed the first sign of wilting on eggplant started about 40 days after inoculation and grew worse over time. Wilting first appeared on the lower leaves before spreading to the higher leaves. Vascular darkening was visible across the entire plant from the first stages of infection onward. Isolates of *F. oxysporum* f. sp. *melongenae* FO3 (77.23%) and FO7 (72.36%) were the most virulent against eggplant plants. Whereas the least virulent strains (25.45 and 36.36%, respectively) were recorded by isolates FO5 and FO8. Two pathogenicity tests were conducted for each.

Nirmaladevi (2012) inoculated the fungus ontwenty days old seedlings by using standard root dip method. Conidia of all the isolates were recovered from one week old cultures. Seedlings were removed from the pot trays and shaken to remove the adhering particles and cleaned carefully under tap water. The roots were trimmed with a sterile scissor and were submerged in

the conidial suspension for 30 mins. The inoculated seedlings were transplanted to minipots, 15cm diameter, surface sterilized with 0.1% mercuric chloride. Containing soil and sand 1:1 ratio and incubated in greenhouse where day and night temperatures varied between 25-30 °C with 12h light and 12h dark. The severity of the disease was assessed from 2 weeks of inoculation up to 45 days. Symptoms were recorded according to a scale ranging from 1 to 5. 1. No symptoms, 2. Slight chlorosis, wilting or stunting of the plant, 3. Moderate chlorosis, wilting or stunting of the plant, 4. Severe chlorosis, wilting or stunting of the plant and 5. Death of the plant.

Altinok (2005) used a modified root-dip inoculation technique for pathogenicity experiments that were carried out twice for each isolate. In seedlings of *Solanum melongena* at the six-leaf stage, 74 different *Fusarium* isolates were examined. While control plants were dipped in sterile tap water, injured roots were immersed for 10 minutes in a conidial suspension (1 × 106 conidia mL−1 in sterile water). Seedlings were raised in growth chambers after being put into pots. A wilt index was used to evaluate the severity of the wilt signs on the leaves after three weeks. All of the tested isolates were harmful to eggplant. All of the injected plants died. Leaf chlorosis and necrosis were symptoms seen on the inoculated plants, which were comparable to those in commercial glasshouses. Symptomless control plants were also present.

Montanari *et al*., (2004) evaluated in a melon *Fusarium oxysporum* f.sp. *melonis* (FOM) pathosystem in a climatic cell at 27°C, 12 hours of daylight, and 70-80% relative humidity,140 days after fortification. To achieve a concentration of 105 conidia/ml, a washed conidial suspension of FOM was added to the potting mix after each organic substance had been mixed individually with sand and peat (1:1:1; v/v/v). Nine 5-day-old melon plants were transplanted into the various compost combinations containing the fungal pathogen one week later. As a control, a peat-sand mixture (PS; 2:1; v/v) inoculated with FOM and either supplemented with TA312B2 or not was utilized. For each treatment, four replicates were created. The following scale was used to evaluate wilt symptoms 27 days later: 0 = no disease; 1 = limited local symptoms; 2 = well developed symptoms; 3 = severe wilt or death.

2.7. Host invation and development of disease by the pathogen

At any stage of cotton growth, *Fusarium oxysporum* infects the roots and colonizes the vascular system, resulting in plant wilt and mortality. Wilted cotyledons, clorosis and necrosis of leaves,

defoliation, vascular darkening and plant mortality are all signs of Fusarium wilt. (Zhu *et al.,* 2021)

By making wounds, nematodes help bacteria and fungus spread infection. Wilt is caused by the fungus and bacteria infecting the plant's vascular system and interfering with the movement of water and nutrients which weakens the plants by preventing them from absorbing water and nutrients. Resulting in death of the plant. Wilt complex refers to the interaction of fungi, bacteria, and nematodes and can occur within the same plant. (Akter *et al*., 2021)

Roots are first symptomlessly penetrated by *F. oxysporum*. In the afterwards, it colonizes vascular tissue and causes extreme wilting necrosis and chlorosis of aerial plant portions. . When compared to other *Fusarium* species, isolates of *F. oxysporum* show a high level of host specificity (Husaini *et al*., 2018).

After coming into contact with the root, hyphal branching is set off, which allows *F.oxysporum* to create hyphal swellings and invade the root. Depending on the *F. oxysporum* strain and plant species involved, the fungal hyphae enter plant roots through wounds, epidermal cracks, lateral root emerging points or by directly penetrating the root tip. The apoplast of the root cortex is how hyphae get to the vascular stele. Both pathogenic and nonpathogenic strains colonize the root cortex, but although the initial colonization pattern is similar, the extent and pattern of colonization differs during later stages. In some cases, intracellular growth is noticeable along with local host cell-death, a phenomenon observed more frequently among non-pathogenic strains (Gordon, 2017).

By penetrating through the roots, *Fusarium oxysporum* restricts the transport of nutrients and water in the vessel. Both its macro and micro conidia are engaged in this process. Chlorosis caused by fungi, slight vein clearance on exterior leaves, and drooping with wilting of the leaves. The entire plant dies when the xylem of the stems turns brown (Arfaoui *et al*., 2007).

The pathogen *Fusarium oxysporum* that enters the roots of its host eggplant by feeding on nematodes and entering through natural or intentional wounds. During its invasion, it penetrates into the plant's xylem vessels, which carry water and nutrients from the roots to the plant's crown, and fungal mycelia create toxins as they spread throughout the plant. When the xylem channel is blocked, the plant dies suddenly (Pietro *et al*., 2003).

F. oxysporum enters the vascular system through the root tissues and quickly colonizes in the xylem vessel. As a result, the host plant begins to exhibit symptoms such as upward, inward

rolling, and yellowing of the leaves, which causes the entire plant to wilt that means death of the host plant (Roncero *et al*., 2003).

2.8 Management of Wilt Disease

Fusarium oxysporum, a soil-born pathogen, is responsible for the wilt disease. Crop rotation, the use of various resistant cultivars, the sterilization and solarization of the soil, and the use of fungicides are some of the primary approaches used to manage Fusarium wilt. Fungicide use does not always produce desirable outcomes and may potentially have negative effects on the ecosystem. Moreover, applying chemicals to treat soil infertility is ineffective since favorable conditions encourage the fungus *Fusarium oxysporum* to recolonize the area. Using biocontrol agents and organic amendments, such as spent mushroom substrate, vermicompost, biochar, and chicken manure, soil-borne diseases have recently been treated. There are still issues with using biocontrol agents to suppress disease because there is insufficient information about the cures and the application of appropriate doses for plant pathogen management.

2.8.1. Management through Spent Mushroom Substrate

The substrate that is still there after the entire crop of mushrooms has been harvested is known as spent mushroom substrate. It is abundant in many different microorganisms, including fungus and bacteria that fight off disease. It is simple to gather, biodegradable, safe to use, less expensive to develop, and capable of controlling a variety of soil borne pathogens.

Ocimati *et al*., (2021) envestigated that *P. ostreatus* and other mushroom species' unsterilized mushroom substrates have antifungal properties that are effective against *F. oxysporum* because they include a variety of microorganisms that are hostile to the pathogenic fungi. Even at low concentrations (1% w/v), unsterilized filtrate of *P. ostreatus* substrates effectively inhibited the development of *F. oxysporum*.

Wang *et al.,* (2020) reported that In the pot experiment, the application of the spent *Flammulina velutipes*, *Lentinus edodes*, and *Pleurotus ostreatus* substrates (FV, LE, and PO) considerably decreased the incidence of Fusarium wilt disease by 53.3%, 25.7%, and 37.9%, respectively.

Salim *et al.,* (2017) observed that Significant results were obtained for the IDM of *F. oxysporum* utilizing the spent mushroom compost, chemical approach (carbendazim), and physical method

(soil solarization). The amount of *F. oxysporum* in plants treated with composted spent mushroom and solarized soil significantly decreased.

Adedeji and Aduramigba (2016) described that microorganisms found in the compost from used mushrooms that in a dual culture experiment, were successful in combating *Fusarium* spp. The interactions between the various species and the degree of *Fusarium* spp. inhibition were observed and noted. It was found that non-autoclaved extracts at concentrations of 1%, 5%, 10%, or 15% greatly reduced the pathogen's ability to multiply by 69.7%, 88.1%, 80.3%, or 85.5%, respectively. The concentrations of unsterilized spent mushroom compost significantly reduced the mycelium growth of *F. oxysporum* f. sp. *Lycopersici*, with concentrations of 0.05, 0.10, and 0.15g/ml being significantly higher. For sterilized spent mushroom compost, the highest concentration (0.15g/ml) was recorded that significantly reduced the mycelial growth of *Fusarium* spp.

Salim and Simon (2015) reported that *P. fluorescens* in combination with spent mushroom compost shows maximum reduction of *Fusarium oxysporum* as compared with other treatments.

Montanari *et al*., (2004) stated that the biological control of wilt disease was significantly improved by binary fortification with Ca-Lignosulphonate and TA312B2 after spent mushroom compost shown high suppressive action. They also mentioned that LS includes monosaccharides (20%) and polysaccharides (5%), which can have a direct biocontrol effect against some pathogens and result in a significant reduction in the mycelial growth of *Fusarium oxysporum*.

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2.8.2 Management through vermicompost

Vermicompost can be helpful in preventing plant wilt disease. Wilt diseases are caused by bacteria and fungus that are present in the soil and attack the roots of plants, causing wilting and eventually death. A significant portion of the beneficial microorganisms in vermicompost, such as bacteria and fungi, can inhibit the growth of soil-borne diseases. Vermicompost can be used to amend soil to boost fertility, improve structure, and promote plant development.

Wang *et al*. (2021) reported that a beneficial organic fertilizer, vermicompost can also be used to prevent and manage plant diseases. This is a significant expansion of the use of vermicompost. Vermicompost is an organic fertilizer that can enhance soil structure, fertility,

and disease resistance. For the ecologically responsible biological control of tomato Fusarium wilt, vermicompost application presents an abundance of antagonists. Actinobacteria, Chloroflexi, Saccharibacteria, and Planctomycetes grew more readily under the vermicompost treatment than under the control treatment, reduce the growth of Proteobacteria, Gemmatimonadetes, Firmicutes, Verrucomicrobia, and Cyanobacteria. Furthermore demonstrated that when vermicompost was used, the incidence of Fusarium wilt was 36.5%–73.9% lower than the control treatment.

Wylie and Punja (2021) suggested that Fusarium wilt can be effectively controlled by using aerated vermicompost tea.

Orosz *et al*. (2021) observed many composts and their water extracts have been studied worldwide for their effects on various plant diseases, particularly those brought on by soilborne pathogens. For the management of plant diseases, such as, compost and compost tea were discovered to be an environmentally friendly choice. Many soil-borne plant pathogenic species can be found in the genus *Fusarium*.

Zhang *et al.,* (2020) reported that the treatment of 50% soil+50% vermicompost was the most successful in preventing cucumber Fusarium wilt and enhancing the development, fruit quality, and yield of *F. oxysporum* infected plant specimens. Therefore, by enhancing the soil, vermicompost applied in moderate amounts may reduce the occurrence of cucumber Fusarium wilt and enhance plant development, yield per plant, and fruit quality.

Geroche, (2019) reported that The control of Fusarium wilt cannot be done using just one technique. Recently, great attention has been paid to a variety of microbiological agents and organic fertilizers for the long-term control of the condition. For instance, the utilization of vermicompost tea and efficient microorganisms that could control wilt disease. 45% of the Fusarium wilt may be controlled by VT (62.50ml) + EM (40ml).

Barman *et al*., (2013) reported that the organic matter resources found in vermicompost are abundant and have the singular potential to enhance the chemical, physical, and biological properties of soils or growing media. According to Garcia, adding vermicompost increases the amount of organic matter and nutrients in the soil, which improves soil productivity and other soil properties. Moreover, they may have strong suppressive effects against infections carried on by a range of soil-borne plant pathogens when used as soil additives. Certain plant diseases like *Pythium* and *Fusarium* as well as nematodes may be suppressed by an increase in the population of particular microbes. The actions of hostile microorganisms have been linked to

the disease suppressive effects of vermicompost. Several of the organic substances used to make potting media were suppressive of Fusarium wilts.

Szczech and Magdalena (2008) reported that vermicompost added to different container media dramatically reduced *Fusarium oxysporum* infection of tomato plants. In direct proportion to the rate of vermicompost treatment, the protective effect grew. Vermicompost extracts that had been sterilized and applied to potato dextrose agar that encourage the growth of *F. oxysporum* mycelium.

2.8.3 Management through biochar

A type of charcoal called bio-char is added to soil as a soil amendment. It is made from a variety of organic materials, such as crop wastes or residues, woodchips, and municipal or urban waste, by pyrolysis, which is a thermal combustion that occurs at temperatures ranging from 200°C to 900°C and in an oxygen-limited environment.

Podeva *et al*., (2021) described the significant percentage of organic matter in biochar increases the calcium, nitrate, phosphorus, and potassium content of the soil. Possess greater water holding capacity and pH as well. Also, it was observed that the combination of compost and green waste biochar (at concentrations ranging from 0 to 3% wt/wt) has beneficial effects in minimizing Fusarium wilt, which operate either directly through antagonism or indirectly through the induction of systemic resistance in plant.

Akanmu *et al.,* (2020) described the capability of biochar to reduce greenhouse gas emissions, improve soil quality, and boost agricultural yield has been investigated. Moreover, biochar has been said to be useful in preventing some plant diseases that are transmitted by the soil and the air. Also reported biochars made from sawdust and poultry manure were successful in treating corn with Fusarium ear disease.

Singh and Kumar (2020) observed that *Fusarium* spp. growth rate in soils treated with bio-char has been decreased to 50% as opposed to the control where the pathogen is growing at a rate of 93%. The Bio-char decreased the chlamydospore growth in the soil, lowering the prevalence of infection carried on by *Fusarium* spp.

Akhter *et al.,* (2016) stated that as chlamydospores are the primary source of inoculum, biochar is effective against a variety of foliar and soil-borne phytopathogenic bacteria and fungi. Because of this, both growers and phytopathologists are very interested in how biochar affects root exudation and subsequent disease suppression phenomena. The ability of biochar and

compost in the plant growth medium to lower the infectivity potential of wilt-inducing chlamydospores was discovered in the study. They claimed that combining green waste biochar and compost as a soil organic amendment favorably stimulated tomato plant development and reduced the capacity of chlamydospores to cause the disease Fusarium wilt.

Khalifa and Thabet (2015) observed that application with total biomass increased by roughly 20% (non-infested controls), 93% (*F. oxysporum* infected plants), and 75% (*R. solani* inoculated plants) in comparison to the corresponding controls, biochar significantly improved tomato plant development. They found that plants growing in amended substrate had higher levels of resistance to *R. solani* and *F. oxysporum*.

Elmer and Pignatello (2011) reported that Coconut charcoal amendments reduced Fusarium crown and root rot, boosted arbuscular mycorrhizal colonization of asparagus seedlings, and produced the plant hormone ethylene, which promotes growth. Less than 2 ppm of ethylene can promote AM germination and hyphal development.

2.8.4 Management through poultry manure

The waste products of poultry grown for meat, eggs, or other agricultural reasons are known as poultry manure. It is frequently used as a fertilizer in agriculture and is a valuable source of nutrients for plants. The three basic minerals for plant growth—nitrogen, phosphorus, and potassium—are abundant in poultry manure. Other micronutrients like calcium, magnesium, and sulfur are also present. High quantities of nitrogen found in poultry manure can encourage plant growth and increase their resistance to Fusarium wilt. However, using too much poultry manure as a fertilizer can also encourage the development of other soil-borne diseases, so it's vital to use it carefully and adhere to recommended application rates.

Islam *et al*., (2021) reported that the treatment of organic amendment, such as trichocompost, chicken manure, etc., has a substantial impact on the incidence of wilt disease. By treating poultry waste, the incidence of the banana pandemic in Gazipur and Ishurdi was reduced to 40% and 41.66%, respectively, and a yield of 29.3 t/ha was noted.

Yasmin *et al*., (2018) reported that the treatments were combined to perform integrated management of fusarium wilt of gladiolus, and it was evident from the results that adding poultry manure at a rate of 5 tons per hectare to the soil 25 days prior to sowing, along with corm treatments at a concentration of 0.1% for 15 minutes, and soil drenching at a concentration

of 0.1% 45 days after sowing, helped improve disease control and increased spike germination, number, and quality.

Jat *et al*., (2017) observed that with a seed yield of 398.89 kg/ha, poultry manure was shown to be the least efficient at decreasing the incidence of Fusarium wilt, controlling the disease up to 41.15 percent under field conditions.

Melero-vara *et al*., (2011) reported that Poultry manure and other N-rich organic amendments suppress the viability of certain soil-borne plant diseases. Final disease occurrences were lower on plots which treated with poultry manure, methyl bromide, or soil solarization.

Islam *et al*., (2010) observed that use of various soil amendments, such as poultry waste, minimized the occurrence of wilt at different days after transplanting. Poultry waste promotes in higher tomato production while Trichoderma harzianum, vermicompost, and poultry waste all demonstrated improved performance against the wilt disease.

Youssef, (2007) reported that the phytopathogen *Fusarium oxysporum*, which causes Fusarium wilt on tomato plants, can be biologically controlled by using composted chicken manure. CMs can prevent soil-borne diseases since both the fungi and the bacterial microorganisms that were isolated from the CMs showed their potential biocontrol action against *F. oxysporum*. Also showed that during an in vitro experiment, CM bacteria also demonstrated their capacity to inhibit *Fusarium* growth.

MATERIALS AND METHODS

A research experiment was conducted to evaluate the effect of spent mushroom substrate and other organic amendment against fungal wilt of brinjal *(Fusarium oxisporum* f. sp. *melonganae*). The selected treatments were evaluated through a field experiment in natural conditions. The details of the methods followed and materials used in the study are described below:

3.1. Experimental site

The field experiment was conducted in the Central Farm of Sher-e-Bangla Agricultural University and lab experiment was conducted in MS laboratory of the Department of Plant Pathology, Sher-e-Bangla Agricultural University, Dhaka-1207.

3.2. Experimental duration

The experiment was carried out during the period from September 2021 to April 2022

3.3. Climate

The experimental site is situated in a subtropical environment which has three different seasons, summer season (March to April), monsoon season (May to October) and winter season (November to February). Plenty of sunshine and moderately low temperature prevails during the experimental period, which is suitable for brinjal production in Bangladesh.

3.4. Soil

The experiment was done in the central farm of Sher-e-Bangla Agricultural University which belongs to the Madhupur Tract (AEZ-28). The site used in this experiment is sited below:

Land type	Medium high land
Field level	Above flood level
Topography	Up land
Drainage	Fairly good
Soil texture	Silty loam
Color	Dark olive-grey color

The physical and chemical characteristics of the soil sample collected from the Soil Resource Development Institute (SRDI), Soil Testing Laboratory, Khamarbari, Dhaka and were presented in Appendix-3

3.5 Variety used

Brinjal variety BARI BT Brinjal 2 (Kajla) was used for the experiment.

Figure 1. Seeds of BARI BT Brinjal 2

3.6 Collection of seeds

Healthy and disease free seeds of BT Brinjal 2 was collected from Bangladesh Agricultural Development Corporation (BADC), Gabtoli, Dhaka.

3.7 Treatments of the experiment

In total 8 treatments with three replications were used to achieve the stated specific objectives. Each treatment combination put once at each block. The treatments were used for this study are given below-

T_0 = Control

T_1 = Spent Mushroom Substrate (SMS) (25 t/ha)

T_2 = Vermicompost (10t/ha)

T_3 = Poultry manure (5 t/ha)

T_4 = Biochar (20 t/ha)

T_5 = Spent Mushroom Substrate + Biochar (25 t/ha + 20 t/ha)

T_6 = Spent Mushroom Substrate + Poultry manure (25 t/ha + 5 t/ha)

T_7 = Spent Mushroom Substrate + Vermicompost (25 t/ha + 10t/ha)

3.8 Collection of materials used as treatment

Spent mushroom substrate collected from National Mushroom Development and Extension Center, Savar, Dhaka. Biochar collected from CCDB biochar project, Manikganj. Poultry manure collected from a poultry farm, Mohammadpur. Vermicompost collected from SAU farm.

3.9. Land preparation

At first ploughed the experimental field with power tiller then mixed with cowdung for the preparation of the experimental field and after which the test area was exposed to sunlight for seven days. Ploughed and cross ploughed the land by using a a country plough for good tilth. After each ploughing, laddering the soil and breaking up the clods into smaller pieces was done. After that weeds were removed from the experimental field. Then plots and drains were made by using a spade.

3.10. Fertilizer and manure application

Name	Application rate(kg ha^{-1})
Cowdung	10,000
TSP	125
Urea	150
MP	100

The total amount of Cowdung and TSP were used during the final land preparation. The application of Urea and MP were done in two installments. First application was done after 15 days of seedlings transplant and second was given at the mid of harvesting.

3.11 Design and layout

The design of the experiment was laid out in Randomized Complete Block Design (RCBD) with eight treatments and three replications. The experimental field was divided into three blocks. Each block had eight plots. As a result, three blocks had 24 plots. The size of each plot was (2.5 x 1.8) m. The space was 0.75 between the blocks and 0.50 was kept between the plots. Each plot had 6 plants and plant to plant distance was 75cm.

Figure 2. Layout Preparation

3.12 Preparation of seedlings

Seedlings were grown with intensive care in the experimental field of SAU farm. At first seedbed was prepared by mixing with Furadan 5G then covered with polythene to sterilize the soil. For the seedlings preparation, seeds were soaked overnight in water and then seeds were sown in the seedbed. Proper care was taken for the germination of seeds and development of the seedlings.

Figure 3. Germination of seedlings

Figure 4. Seedlings ready to transplant

3.13 Application of treatments

The chosen treatments were applied to the main field 20 days before transplanting the seedlings to ensure adequate decomposition, the growth of combative microorganisms and the development of the pathogen suppressing abilities.

3.14 Transplanting of seedlings

After 25 days of germination seedlings were prepared to transplant in the main field. Firstly, the seedbeds were watered before uprooting the seedlings to minimize the root damage. Transplanting was done in the afternoon. Seedlings were transplanted in the main field on 7 December 2021. Plant to plant distance was maintained 75cm and each plot had 6 plants. Sufficient irrigation was given just after the transplanting to keep seedlings upright.

Figure 5. Transplanting of seedlings

3.15 Intercultural operations

After transplantation the field was kept under careful observation. Various intercultural operations. eg. Irrigation, gap filling, weeding, earthing up, fertilizer application were done during the experimental period for the better production of crop.

3.15.1. Gap filling

If any plant died gap filling was done after 7days of seedlings transplanting. Seedlings were collected from the same source. Gap filling was done only where it was necessary.

3.15.2. Irrigation

When necessary irrigation was given throughout the growing season. Because irrigation is necessary to keep the soil moistened. After weeding and fertilizer application flood irrigation was given and excess water was allowed to be drained out.

3.15.3. Weeding

After 15 to 20 days of transplanting weeding was done to keep the field free from weeds and debris. Weeding was done at every 15 days interval from planting to flowering stage. The experimental field were observed regularly. Field sanitation was maintained throughout the growing season. Infected plants, blighted leaves, wilted and dead plants were removed regularly to keep the field clean.

3.15.4. Earthing up

When needed earthing up was done during the experimental period.

3.15.5. Insect pest control

Actara 2gm per litter was applied as preventive measure to reduce the attack of insect pest eg. cutworm, leafhopper etc in the experimental field. Different cleaning practice was done to reduce the insect pest attack. Removed infected plants and debris to reduce insect pest.

3.16. Harvesting

Harvesting was started at 4 march 2022. During the harvesting period at each harvest, the number of fruits, individual weight of fruit, total weight of the fruits and individual fruit diameter was taken plot wise and was kept the data in a note book.

3.17. Collection of data

The following parameters were considered for data collection.

Observations: (Disease incidence)

- Number of infected plant/plot
- % incidence

Observations: (yield and yield contributing characters)

1. Plant height (cm)
2. Number of branches /plant
3. Number of leaves /plant
4. Number of fruits /plant
5. Number of fruits/plot
6. Fruit length
7. Individual weight of fruits
8. Yield /plant
9. Yield /plot
10. Yield /ha

3.17.1. Plant height (cm)

Plant height was measured with a measuring tape.

3.17.2. Number of branches /plant

Branches were counted and collected data in the note book.

3.17.3. Number of leaves /plant

Number of leaves /plant were counted and entry the data.

3.17.4. Number of fruits /plant

Harvesting was done at 10 to 15 days interval. Counted the fruit /plant and collected data in a note book during each harvest period. The sum of the all harvested fruits per plant from 1st harvesting day to last harvesting day was the fruits/ plant.

3.17.5. Fruit length

Harvested fruits were measured horizontally by using a measuring tape to obtain the fruit length.

3.17.6. Individual Fruit weight

By using a weight machine individual fruit weight was measured.

3.17.7. Yield /plot

Weight of all fruits /plot from the 1st harvesting day to last harvesting day was the yield /plot.

3.17.8. Yield /plant

Weight of fruits/plot were measured by using a weight machine and collected the data. Then when the last harvest was done sum the total weight of fruits/plot. Then used the following formula to get yield/plant.

$$\text{Yield per plant} = \frac{\text{Sum weight of all fruits/plot}}{\text{Total number of plants/plot}}$$

3.17.9. Number of infected plant/plot

The experimental field was observed regularly. Counted the infected plants/plot and collected the data in a note book.

3.17.10. Disease incidence (DI) (% plant infected)

On the basis of infected plant, disease incidence was measured in percentage. Incidence of wilt disease were recorded by observation of visual symptoms at 25, 45, 65 days after transplanting. Firstly count was done how many plant were infected per plot then applied the following formula to get disease incidence in percent.

$$\text{Disease incidence (\%)} = \frac{\text{Infected plant per plot}}{\text{Total plant per plot}} \times 100$$

3.18. Phytopathological study

3.18.1. Preparation of PDA (Potato Dextrose Agar) media for culturing of *Fusarium oxysporum* f. sp. *melongenae*

200g of sliced, washed, unpeeled potatoes were boiled in 1 liter of distilled water for 30 minutes to make potato extract, which was then decanted via cheesecloth. Then distilled water was added so that the overall suspension had 1 liter volume. Then 20 grams of dextrose and 20 grams of agar powder added. The medium was then sterilized by autoclaving at 15 pounds per square inch (15 psi) pressure for 45 minutes.

3.18.2 .Collection of disease sample

Infected plants which showing wilt like symptoms were collected by using a polythyene bag to transport the infected stem with root that had been removed from the experimental field to the lab of the Department of Plant Pathology at Sher-e-BangIa Agricultural University, Dhaka-1207 for primary examination. The specimen diseased samples were rinsed with running tap water to dispel adherent soil particles and then squeezed in between the fold of aseptic blotting paper to remove excess water and preserved at 4-6°C in refrigerator for further analyses. The entire specimens were examined in the laboratory for the presence of the causal organism.

3.18.3. Isolation of *Fusarium oxysporum* f. sp. *melongenae* from symptomatic brinjal plant

Diseased stem was collected from the experimental field and then the diseased stem was taken to the laboratory of the Department of Plant Pathology, Sher-e-Bangla Agricultural University, Dhaka-1207. After that the vascular portion of the diseased stem was cut into small pieces (.05-1cm) by a sterilized knife and each piece was having small bits of disease and healthy tissues. Sterilization was done by dipping in 0.01% mercuric chloride ($HgCl_2$) for 2–3 minutes. By using a sterilized forcep the chopped pieces then placed on a blotter paper after 3 times washed with distilled water and then incubated at 25±1°C for 7–10 days. When the fungus was grown then transferred it on PDA media. The pathogen was then purified by the transfer of mycelium

from the tip of the colony to another petri plate at least three times which was previously poured with sterilized PDA in aseptic condition. This method was followed by Das (2021) and later by using a compound microscope the pathogen was identified as *Fusarium oxysporum* f. sp. *melongenae.*

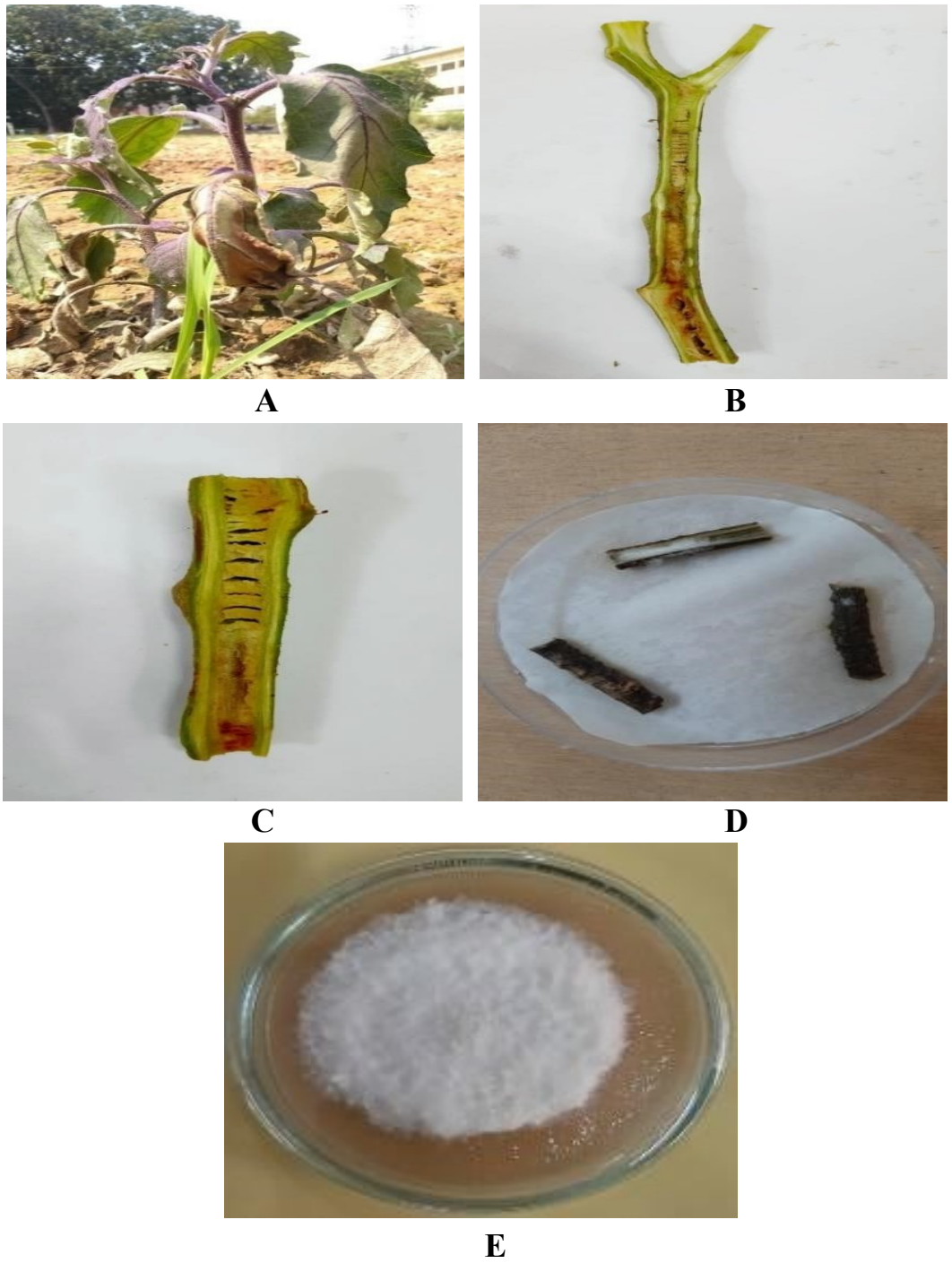


Plate 1. Isolation of *Fusarium oxysporum* f. sp. *melongenae* A. Infected plant B. Vascular discoloration C. Small piece of infected stem D. Pathogen on blotter paper E. Pathogen on PDA media

3.19. Pathogenicity test under pot culture

3.19.1. Soil sterilization and pot preparation

The soil was taken from the field. The clods were broken and the stables were removed. After that, 0.4% formalin solution was completely mixed with soil @ 200ml/cft soil and kept under polythene sheet for 48 hours to retain the gases within the soil for sterilization. The soil was then exposed to sunlight for seven days. After 7 days the treated soil was ready to use. The soil was then placed in 25 cm diameter surface sterilized pots.

Figure 6. Soil sterilization

3.19.2. Seedling preparation

BT Brinjal 2 seedlings were raised in plastic pot. Sterilized soil having fertilizers as per the package of practices was used for Seedbed preparation. The seedlings were watered and checked on a regular basis.

Figure 7. Raising of seedlings

3.19.3. Pathogenicity test of wilt of brinjal

Thirty days old seedlings of brinjal were treated with spore suspension of *F. oxysporum* by root dip method. Damaged roots were immersed in a conidial suspension (106 conidia/ml) for 10 minutes while the control plants were plunged in sterile tap water. After that, seedlings were placed into clean pots. The plants were watered regularly and observed for appearance of wilt symptoms. Observations were done on wilt symptoms for up to 5 weeks .After three weeks of inoculation, symptoms were seen on the inoculated plant, and the pathogen was reisolated and compared with the original culture of *F. oxysporum* to satisfy the Koch's postulates. This method was followed by Das (2021).

3.20. Analysis of data

The data were collected from the experimental field that were statistically analyzed by using computer-based software Statistix 10 software. The data were analyzed by using analysis of variance (ANOVA) to find out the variation of results from experimental treatments.
Treatment means were compared by LSD.

RESULTS

This chapter presents the findings from the current study on the effectiveness of eight different treatments with spent mushroom substrate, vermicompost, poultry manure, biochar, spent mushroom substrate + vermicompost, spent mushroom substrate + poultry manure, spent mushroom substrate + biochar and control for the management of Fusarium wilt of brinjal. The effectiveness of the treatments was evaluated based on many factors, including wilt incidence, plant growth traits, and yield of the crop.

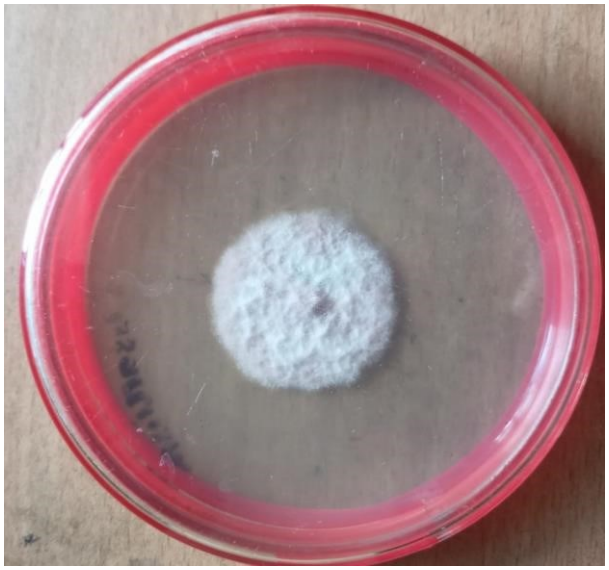
4.1 Isolation and identification of causal agent

4.1.1. Isolation of the pathogen

For isolation and identification of the pathogen the vascular portion of the diseased stem was chopped into small pieces (0.5-1 cm) and then surface sterilized by dipping in 10% solution of sodium hypochlorite for 2–3 minutes, or 0.01% solution of $HgCl_2$ for 30 seconds. With the use of sterile forceps, the chopped pieces were washed three times in water before being placed onto PDA media. (Appendix-5) in a sterilized petridish and incubated at 25±1°C for 7– 10 days. By using hyphal tip culture method the pathogen was purified and grown on PDA media at 25±1°C for 2 weeks and finally the pure culture was preserved at 4°C in refrigerator for future study (Das 2021; Ram *et al*., 2022). Then the pathogen was identified as *Fusarium oxysporum* f. sp. *melongenae* by a compound microscope.

a. Two days old culture

b. Three days old culture



c. Five days old culture

Plate 2. Photographs showing isolation of the pathogen

4.1.2. Identification of the pathogen

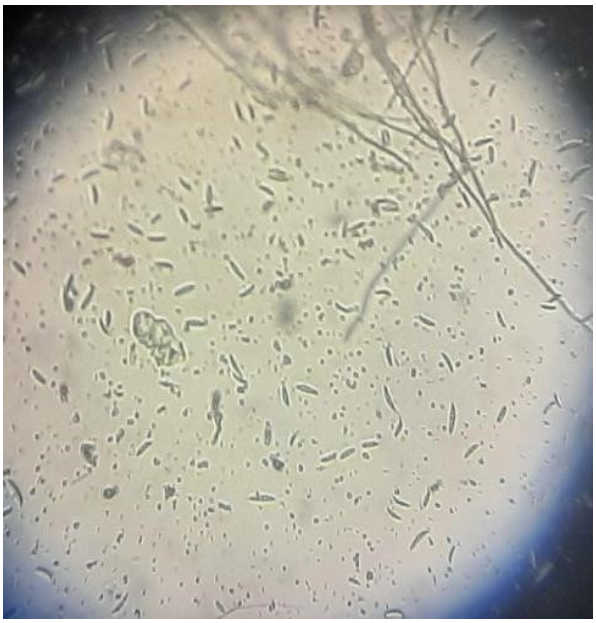
The pathogen was identified under a compound microscope based on symptomatology by mounting spores on a slide, and the pathogen was identified based on morphological traits such as colony growth, color (purple and white), and conidiophores with short microconidia. (Soesanto *et al*., 2011).The fungus flourished in PDA with whitish and light pink mycelium that eventually expanded into a light gray colony as a result of sporulation. By using a compound microscope it was observed that in pure culture, the pathogen developed 2 or 3 celled, slightly curved macroconidia and single cell microconidia. This result is similar to Hasan (2020) and Adedeji *et al*.,(2016). Based on the available findings, the fungus was identified as *Fusarium oxysporum* f. sp. *melongenae.*

a. Whitish mycelium of *F. oxysporum*

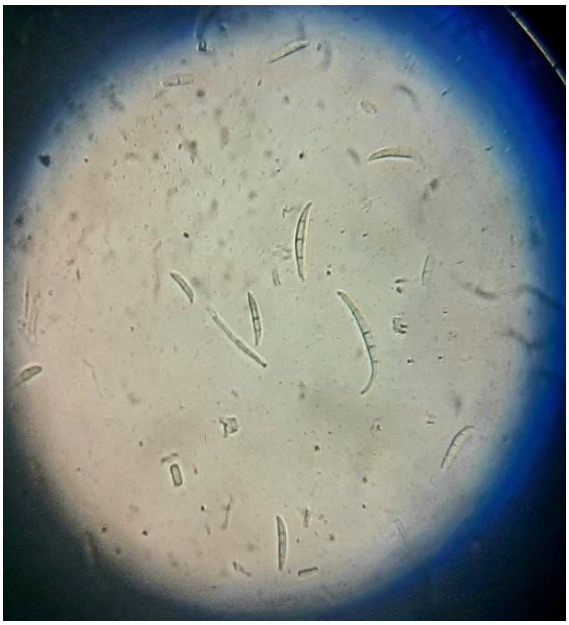
b. Light gray colony of *F. oxysporum*

c. White mycelium of *F. oxysporum*

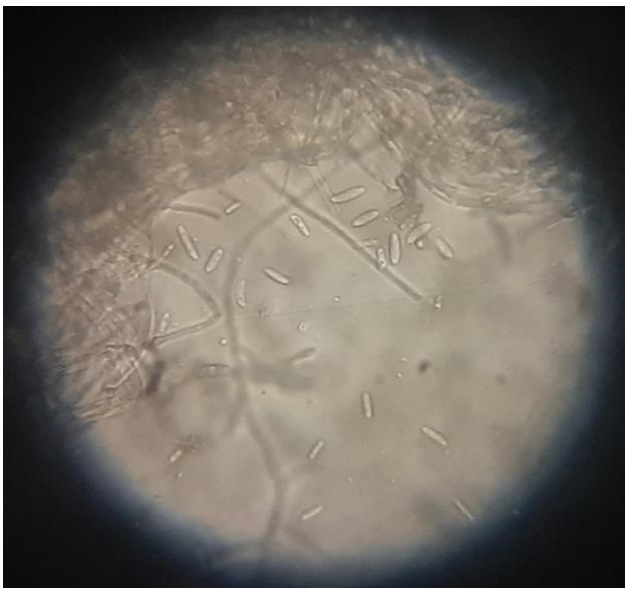
Plate 3. Pure culture of *Fusarium oxysporum* f. sp. *melongenae* on PDA media



a. Conidia of *F. oxysporum* f. sp. *melongenae*



b. Macroconidia of *F. oxysporum*



c. Microconidia of *F. oxysporum*

Plate 4: Identification of *F. oxysporum* f. sp. *melongenae*

4.2. Pathogenicity test of *F. oxysporum* f. sp. *melongenae*

The causal agent of wilt of brinjal (*F. oxysporum* f. sp. *melongenae*) was inoculated into the brinjal plant using the root dip method to investigate the fungus either pathogenic or nonpathogenic. Wilt symptoms were caused for up to 5 weeks. The injected plant showed typical wilting symptoms. Begum (2007) discovered that *F. oxysporum* f. sp. *lycopersici* was capable of producing wilting symptoms in tomato plants, which supported the outcome. The findings were also consistent with those of Altinok (2005), who observed wilting in eggplants of Turkey caused by *F. oxysporum* f. sp. *melongenae*. Reisolation of *Fusarium oxysporum* f. sp. *melongenae* was successfully done from the stem of disease affected inoculated plant, thereby completing Koch's postulates.

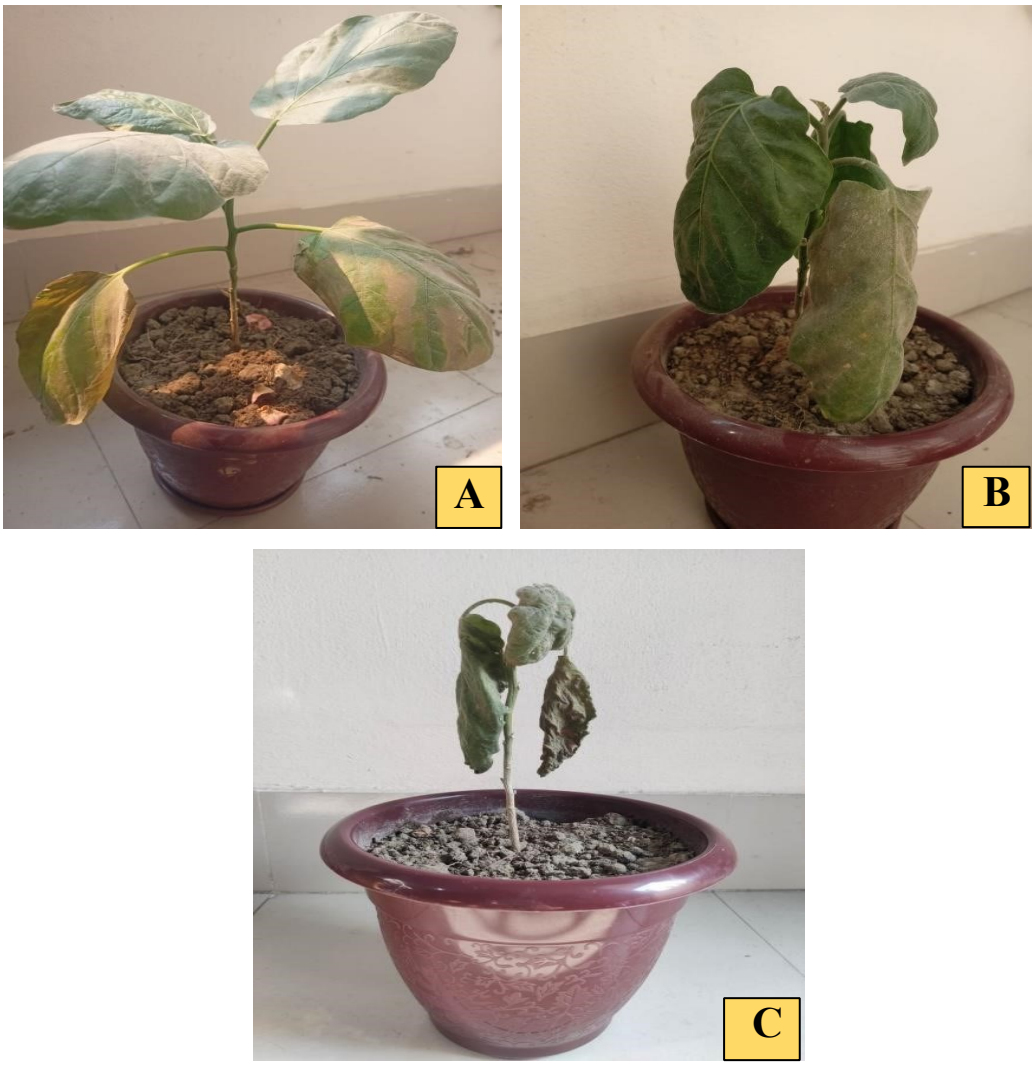


Plate 5. Pathogenicity test of *F. oxysporum* f. sp. *melongenae;* A. Healthy brinjal plant, B. Pathogen inoculated, C. wilted plant due to disease occurance

4.3 Effect of the treatments on number of infected plant and wilt incidence at different Days After Transplanting (DAT)

Based on the visible symptoms, the effect of several treatments on the disease incidence (%) of fusarium wilt of brinjal was observed. Disease incidence (%) was measured three times at 25, 45 and 65 days after transplanting (DAT).

4.3.1. Effect of the treatments on number of infected plant and wilt incidence at 25 Days After Transplanting (DAT)

The treatments used to treat brinjal wilt differed considerably in terms of disease-related parameters at 25 days after transplanting (Table 1). In the field study, the effect of different treatments on the number of infected plants and disease incidence of fusarium wilt of brinjal was observed. After 25 days of transplanting, the number of infected plants was lowest (0.33) for T_5 (Spent Mushroom Substrate + Biochar) and the highest was 2.66 for T_0 (control), which was statistically identical to T_7 (Spent Mushroom Substrate + Vermicompost) (2.00). The second lowest infection rate (0.66) was recorded from T_1 (Spent Mushroom Substrate), T_2 (vermicompost), and T_4 (Biochar) showed statistically similar values. The impact of T_3 poultry manure treatments on the number of infected plants (1.66) differed significantly from the untreated control.

When compared to the control, every treatment greatly decreased the incidence of fusarium wilt and it was 44.4% which was statistically equivalent to T_7 (33.33%). Followed by T_3 (27.77%). The lowest incidence of disease (5.55%) was recorded from T5 (combination of biochar and sms). T_6 (spent mushroom substrate+ poultry manure) showed the second-lowest incidence of disease (11.11%). Incidence of disease (16.66%) was significantly similar in T_1, T_2 and T_4.

Table 1. Effect of different treatments on number of infected plant and the wilt incidence of brinjal at 25 Days After Transplanting (DAT)

Treatments	Number of infected plant per plot	Disease Incidence (DI %)
T_0	2.66 a	44.44 a
T_1	1.00 b-d	16.66 b-d
T_2	1.00 b-d	16.66 b-d
T_3	1.66 a-c	27.77 a-c
T_4	1.00 b-d	16.66 b-d
T_5	0.33 d	5.55 d
T_6	0.66 cd	11.10 cd
T_7	2.00 ab	33.33 ab
CV (%)	58.83	58.83

T_0 = Control ; T_1 = Spent Mushroom Substrate (SMS) ; T_2 = Vermicompost ; T_3 = Poultry manure ; T_4 = Biochar ; T_5 = Spent Mushroom Substrate + Biochar ; T_6 = Spent Mushroom Substrate + Poultry manure ; T_7 = Spent Mushroom Substrate + Vermicompost

4.3.2. Effect of the treatments on number of infected plant and the wilt incidence at 45 Days After Transplanting (DAT)

4.3.2.1 Number of Infected plants

After 45 days of transplanting, the effect of different treatments on the number of infected plants and disease incidence of fusarium wilt of brinjal was observed and the result shows in table 2. The lowest number of infected plants (1.0) was observed from T_5 (Spent Mushroom Substrate + Biochar) followed by T_1 (1.33) and T_6 (1.66) and highest (3.33) for T_0 (control), which was statistically identical to T_2, T_4 and T_7 (2.33). The result of T_2 (Vermicompost) and T_7 (SMS+ Vc) were statistically identical.

4.3.2.2 Wilt Disease Incidence

Fusarium wilt disease incidence varied from 55.55- 16.66% in present study. T_2 (vermicompost), T_4 (Biochar) and T_7 (Spent Mushroom Substrate + Vermicompost) showed the statistically identical result and it was 38.88%. The lowest disease incidence was recorded in T_5 (spent mushroom substrate + biochar) whereas the highest incidence was observed in T_0 (control). The statistically similar results were found in T_1 (22.77%), T_3 (33.47%) and T_6 (27.77%).

Table 2. Effect of different treatments on the wilt incidence of eggplant at 45 Days After Transplanting (DAT)

Treatments	Number of infected plant per plot	Disease Incidence (DI %)
T_0	3.33 a	55.55 a
T_1	1.33 bc	22.22 bc
T_2	2.33 ab	38.87 ab
T_3	2.00 bc	33.33 bc
T_4	2.33 ab	38.87 ab
T_5	1.00 c	16.66 c
T_6	1.66 bc	27.77 bc
T_7	2.33 ab	38.87 ab
CV (%)	35.98	35.99

T_0 = Control; T_1 = Spent Mushroom Substrate (SMS) ; T_2 = Vermicompost ; T_3 = Poultry manure ; T_4 = Biochar ; T_5 = Spent Mushroom Substrate + Biochar; T_6 = Spent Mushroom Substrate + Poultry manure; T_7 = Spent Mushroom Substrate + Vermicompost

4.3.3. Effect of the treatments on number of infected plant and the wilt incidence at 65 Days After Transplanting (DAT)

The effect of different treatments on the number of infected plants and disease incidence of fusarium wilt of brinjal was observed and 65 days after transplanting the results displays in Table 3 .The lowest number of infected plants (1.0) were observed from T_5 (Spent Mushroom Substrate + Biochar) and the highest number of infected plants (4.66) were observed from T_0 (control).The second lowest infected plants (1.33) were recorded from T_6 (Spent Mushroom Substrate + poultry manure) which was statistically similar with T_1 (Spent Mushroom Substrate).The other treatments T_2 (vermicompost), T_3(poultry manure) and T_4 (Biochar) and T_7 (Spent Mushroom Substrate + vermicompost) have statistically similar values.

The highest disease incidence (77.77%) was recorded in control (T_0) and the lowest disease incidence (16.66%) was recorded in T_5 (spent mushroom substrate + biochar). The second lowest disease incidence (27.77%) was recorded in T_6 (spent mushroom substrate + poultry manure) which was statistically similar with T_1 (Spent Mushroom Substrate) (33.33%). T_2

(vermicompost) (49.99%), T_3 (poultry manure) (49.99%), T_4 (Biochar) (44.44%) and T_7 (Spent Mushroom Substrate + vermicompost) (44.44%) have statistically similar values.

Table 3. Effect of different treatments on the wilt incidence of eggplant at 65 Days After Transplanting (DAT)

Treatments	Number of infected plant per plot	Disease Incidence (DI %)
T_0	4.66 a	77.77 a
T_1	2.00 bc	33.33 bc
T_2	3.00 b	49.99 b
T_3	3.00 b	49.99 b
T_4	2.66 b	44.44 b
T_5	1.00 c	16.66 c
T_6	1.66 bc	27.77 bc
T_7	2.66 b	44.44 b
CV (%)	35.96	35.97

T_0 = Control ; T_1 = Spent Mushroom Substrate (SMS) ; T_2 = Vermicompost ; T_3 = Poultry manure ; T_4 = Biochar ; T_5 = Spent Mushroom Substrate + Biochar ; T_6 = Spent Mushroom Substrate + Poultry manure ; T_7= Spent Mushroom Substrate + Vermicompost

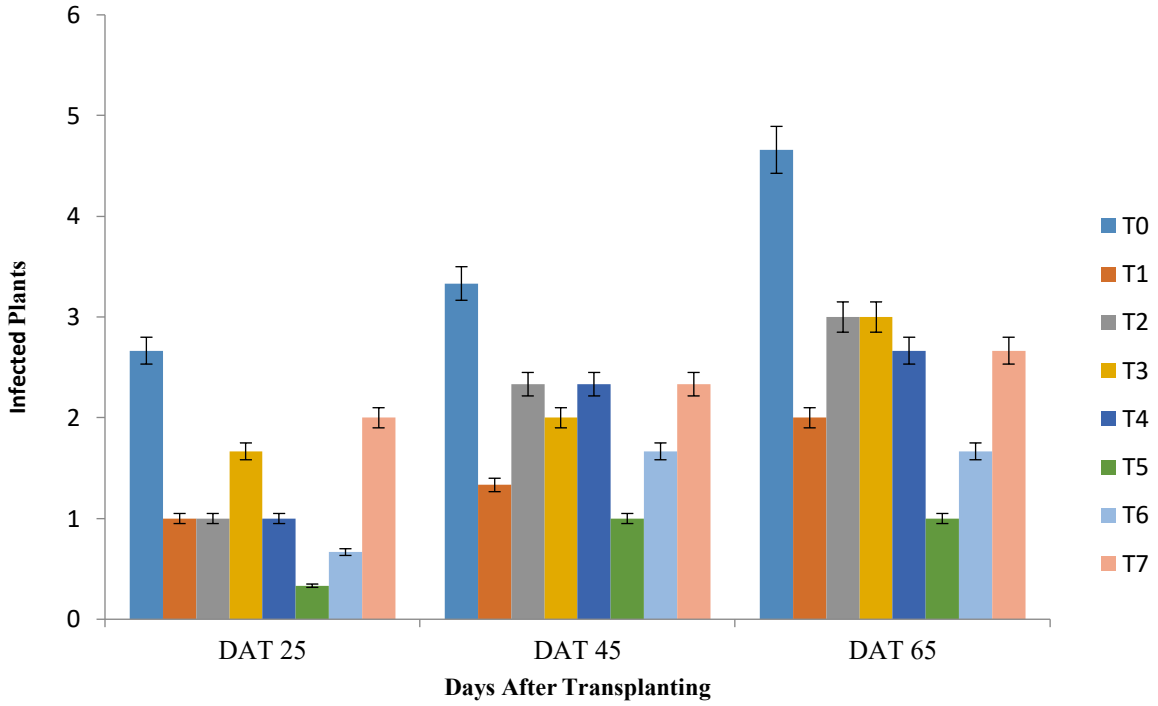


Figure 8. Effect of different treatments on the No. of Infected Plants recorded from 25 DAT to 65 DAT with 20 days intervals.

4.4. Effect of different treatments on plant growth parameters of brinjal

Regarding plant growth indicators, the treatments used to treat brinjal wilt differed significantly and the result shows in Table 4.

4.4.1. Plant height (cm)

In case of plant height, the maximum plant height (71.0 cm) was observed in T_5 (Spent Mushroom Substrate + Biochar) whereas the minimum plant height (50.66cm) was recorded in the T_0 (control) which was statistically similar with T_4 (52.33 cm) and T_1 (54.66 cm). The second highest plant height (66.33 cm) was recorded in T_6 (Spent Mushroom Substrate + Poultry manure). Plant height was recorded from T_2 (Vermicompost) (58.33 cm), T_3 (Poultry manure) (62.0 cm) and T_7 (Spent Mushroom Substrate + vermicompost) (60.33 cm) that was differed significantly from the untreated control.

4.4.2. Number of leaves per plant

In case of number of leaves per plant, the highest number of leaves per plant (111) was recorded in T_5 (Spent Mushroom Substrate + Biochar) and the lowest number of leaf per plant (73.67) was recorded in T_0 (control). The second highest number of leaves per plant (101.67) was found

in T_6 which was statistically similar with T_3 (98.33). The third best result was found in T_1 (89.33) .T_2 (86.0) and T_4 (85.0) statistically showed similar values. T_7 (Spent Mushroom Substrate + vermicompost) (82.0) showed the result that differed significantly from the untreated control.

4.4.3. Number of branches per plant

In case of number of branches per plant, the highest number of branches per plant (11.33) was recorded in T_5 (Spent Mushroom Substrate + Biochar) which was statistically similar with T_6 (10.66) and the lowest number of branches per plant (5.67) was recorded in T_0 (control). The second highest number of branches per plant (8.66) was found in T_3 (poultry manure). T_1 (7.66), T_2 (7.33), T_4 (7.0) and T_7 (7.33) have statistically similar result that was differed significantly from the control.

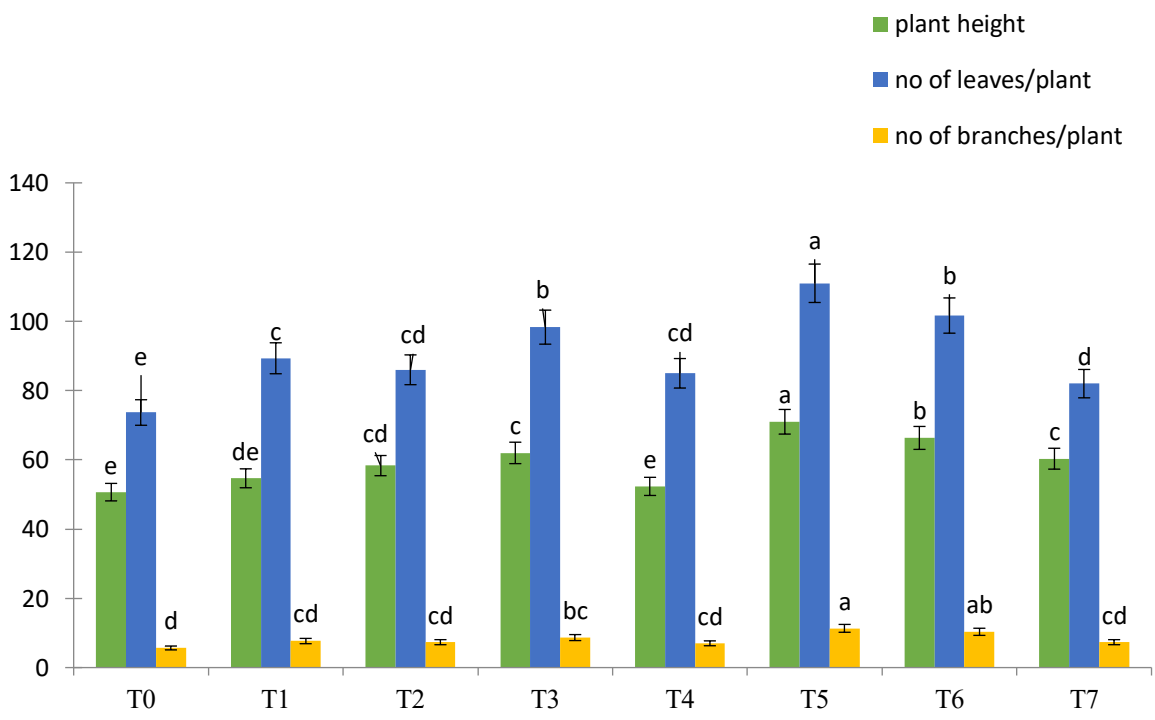


Figure 9. Effect of different treatments on plant growth parameters of brinjal

4.5. Effect of different treatments on yield contributing characters

4.5.1. Number of fruits/plant

In case of number of fruits per plant, the highest number of fruits per plant (12.27) was recorded in T_5 (Spent Mushroom Substrate + Biochar) and the lowest number of fruits per plant (3.77) was recorded in T_0 (control). The second highest number of fruits per plant (8.55) was recorded in T_6 which was statistically similar with T_2 (7.77) and T_4 (8.38). T_3 produced (6.66) number of fruits per plant. T_1 (4.77) and T_7 (4.72) showed statistically similar result.

4.5.2. Number of fruits/Plot

In case of number of fruits per plot, the highest number of fruits per plot (73.66) was recorded in T_5 (Spent Mushroom Substrate + Biochar) and the lowest number of fruits per plant (22.66) was recorded in T_0 (control). The second highest number of fruits per plant (51.33) was recorded in T_6 (Spent Mushroom Substrate + Poultry manure) which was statistically similar with T_2 (46.66) and T_4 (50.33). Number of fruits per plot (40.66) produced in T_3 (poultry manure). T_1 (Spent Mushroom Substrate) (28.66) and T_7 (Spent Mushroom Substrate + Vermicompost) (28.33) showed statistically similar results.

4.5.3. Individual weight of fruits

In case of Individual weight of fruit, the highest fruit weight was found in T_5 (92.51 g) that was statistically identical with T_6 (92.09 g). The second highest fruit weight was found in T_4 (81.73g). The third best result was found from T_7 (76.44g) which showed statistically similar result with T_0, T_1 and T_3. The individual weight of fruit 66.69g was recorded in T_2 (Vermicompost).

4.5.4. Fruit length (cm)

In case of Fruit length, the highest fruit length was found in T_6 (9.11cm) that was statistically similar with T_5 (9.01cm). The untreated control T_0 and T_3 have statistically similar result. T_1 (Spent Mushroom Substrate) (8.56), T_2 (Vermicompost) (8.28) T_4 (Biochar) (8.81), T_7 (Spent Mushroom Substrate + Vermicompost) (8.21) also showed statistically similar result.

Table 4. Effect of different treatments on yield contributing characters

Treatments	Number of fruits/plant	Number of fruits/Plot	Individual weight of fruit (gm)	Fruit length (cm)
T_0	3.77 d	22.66 d	73.32 bc	7.84 b
T_1	4.77 cd	28.66 cd	73.87 bc	8.56 ab
T_2	7.77 b	46.66 b	66.69 c	8.28 ab
T_3	6.77 bc	40.66 bc	73.14 bc	7.80 b
T_4	8.38 b	50.33 b	81.73 b	8.81 ab
T_5	12.27 a	73.66 a	92.52 a	9.01 a
T_6	8.55 b	51.33 b	92.09 a	9.11 a
T_7	4.72 cd	28.33 cd	76.44 bc	8.21 ab
CV (%)	19.35	19.33	7.12	7.16

T_0 = Control; T_1 = Spent Mushroom Substrate (SMS); T_2 = Vermicompost; T_3 = Poultry manure ; T_4 = Biochar ; T_5 = Spent Mushroom Substrate + Biochar; T_6 = Spent Mushroom Substrate + Poultry manure; T_7 = Spent Mushroom Substrate + Vermicompost

4.6. Effect of different treatments on yield of brinjal against Fusarium wilt

The treatments applied for the management of Fusarium wilt of brinjal differed significantly in respect of fruit yield. Results are presented in table 6

The highest yield per plant (865g) was recorded in case of T_5 where Spent Mushroom Substrate + Biochar were applied followed by T_4 (Biochar). T_6 (Spent Mushroom Substrate + Poultry manure) (684g), Treatment T_2 produced the third highest yield (599.33g) followed by T_3 (542g).T_1 (Spent Mushroom Substrate) and T_7 (Spent Mushroom Substrate + Vermicompost) produced 353.67g and 364.67g yield per plant respectively. The lowest yield per plant (277.67g) was noted in T_0 (control).

The highest yield per plot ranged from 5.193-1.66kg. T_5 where Spent Mushroom Substrate + Biochar was applied showed the highest whereas the lowest was revealed from T_0. The second highest yield (4.14kg) was recorded from T_4 statistically similar with T_6 (Spent Mushroom Substrate + Poultry manure) (4.10kg), Treatment T_2 produced the third highest yield (3.59kg) followed by T_3 (3.25kg). T_1 (Spent Mushroom Substrate) and T_7 (Spent Mushroom Substrate + Vermicompost) produced 2.12kg and 2.18kg yield per plant respectively. The lowest yield per plant (1.66kg) was noted in T_0 (control).

The yield varied significantly from 12.7-4.08 t/ha in the present study. The highest yield per hectare was recorded in case of T_5 (Spent Mushroom Substrate + Biochar) followed by T_4 (10.12t/ha) and T_6 (10.05 t/ha). In case of T_2 the yield was recorded 8.79 t/ha followed by T_3 (7.95t). T_1 (Spent Mushroom Substrate) and T_7 (Spent Mushroom Substrate + Vermicompost) produced 5.19t/ha and 5.33t/ha yield respectively. The lowest yield per plant (4.08t/ha) was noted in T_0 (control).

Table 5. Effect of different treatments on yield of brinjal against Fusarium wilt

Treatments	Yield (g/plant)	Yield (kg/plot)	Yield (ton/ha)
T_0	277.67 d	1.66 d	4.08 d
T_1	353.67 cd	2.12 cd	5.19 cd
T_2	599.33 b	3.59 b	8.79 b
T_3	542.00 bc	3.25 bc	7.95 bc
T_4	691.00 ab	4.14 ab	10.12 ab
T_5	865.00 a	5.19 a	12.71 a
T_6	684.00 ab	4.10 ab	10.05 ab
T_7	364.67 cd	2.18 cd	5.33
CV (%)	21.97	21.97	22.17

T_0 = Control; T_1 = Spent Mushroom Substrate (SMS); T_2 = Vermicompost; T_3 = Poultry manure; T_4 = Biochar; T_5 = Spent Mushroom Substrate + Biochar; T_6 = Spent Mushroom Substrate + Poultry manure; T_7 = Spent Mushroom Substrate + Vermicompost

4.7 Relationship between fruit per plant and percent disease incidence at 65 DAT

From the relationship study between fruit per plant with percent disease incidence at 85 DAT, it was revealed that all the treatments significantly increased number of fruits per plant with the decreased of percent disease incidence. The highest result was found by the combined application of Spent mushroom substrate and biochar (T_5) which was followed by T_6 (Spent Mushroom Substrate + Poultry manure).

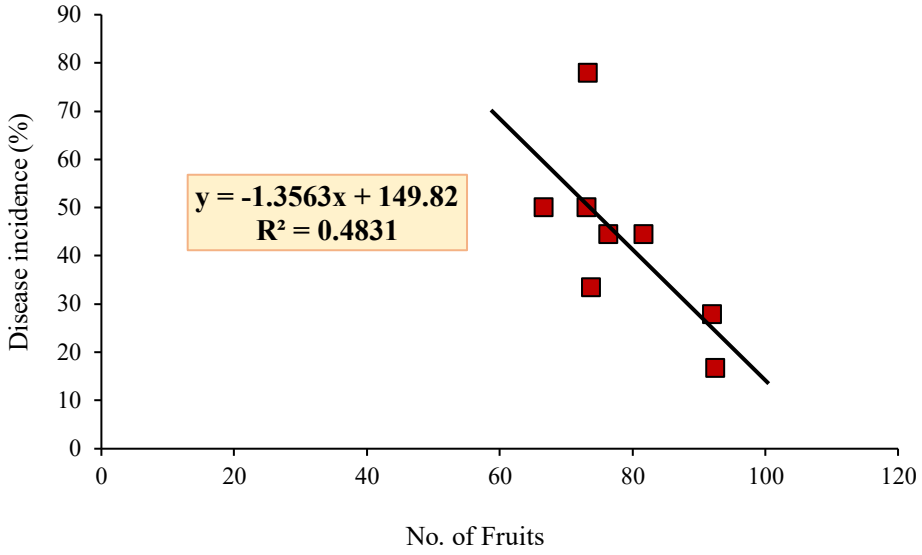


Figure 10. Correlation between fruit per plant and percent disease incidence at 65 DAT

4.8 Relationship between individual fruit weight (gm) and percent disease incidence at 65 DAT

In case of the relationship study between individual fruit weight (gm) with percent disease incidence at 85 DAT, it was revealed that all the selected treatments showed the result that was differed significantly from the untreated control. With the decreased of percent disease incidence individual fruit weight (gm) was increased. The highest result was found in T_5 which was followed by T_6.

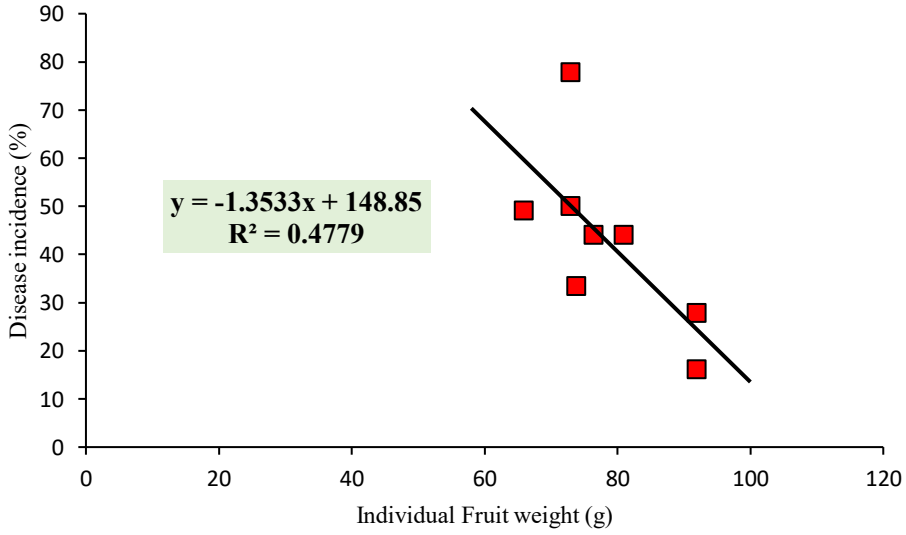
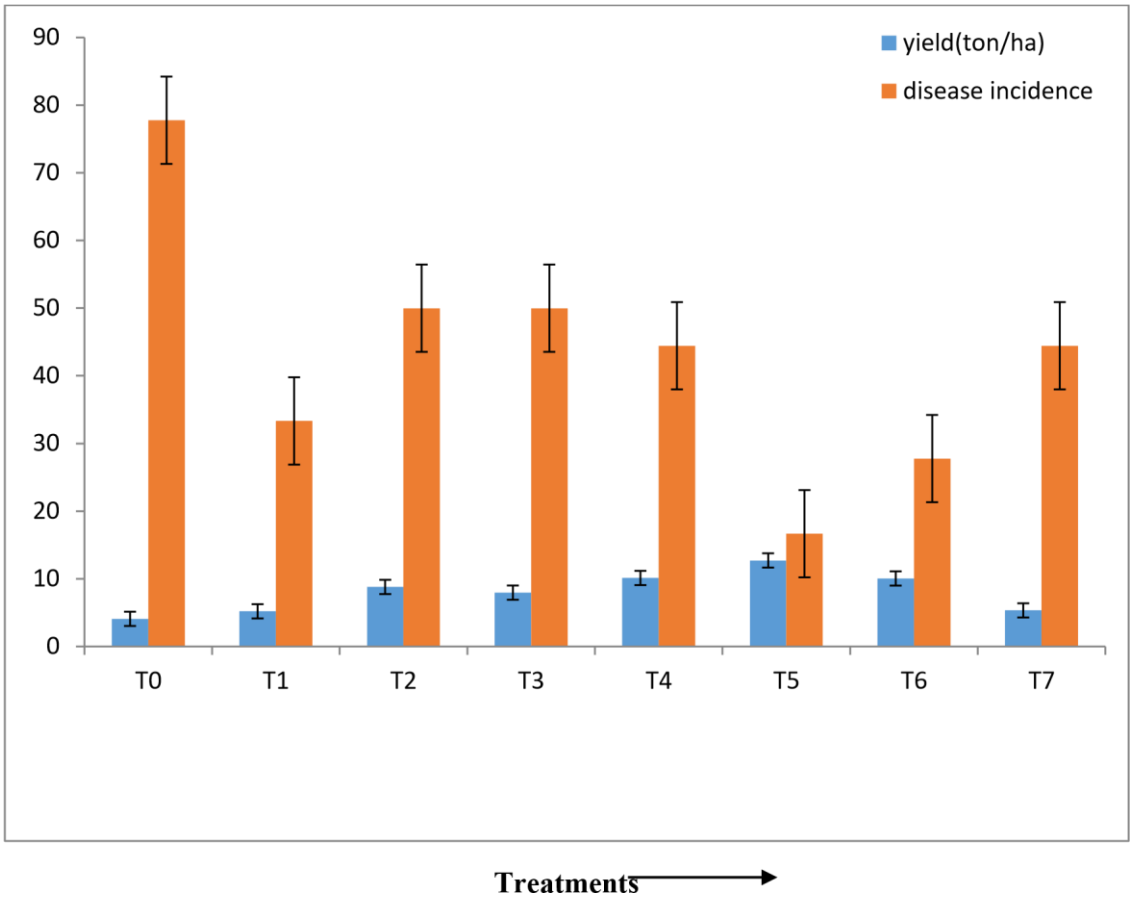


Figure 11. Correlation between individual fruit weight (gm) and percent disease incidence at 65 DAT

4.9 Relationship between yield (ton/ha) and percent disease incidence at 65 DAT

In case of the relationship study between yield (ton/ha) and percent disease incidence at 65 DAT, it was revealed that all the selected treatments showed the result that differed significantly from the untreated control. The yield was increased with the decreased of percent disease incidence. The highest result was found by the combine application of Spent mushroom substrate and biochar (T_5) which was followed by T_6 (Spent Mushroom Substrate + Poultry manure).



T_0 = Control; T_1 = Spent Mushroom Substrate (SMS); T_2 = Vermicompost; T_3 = Poultry manure; T_4 = Biochar; T_5 = Spent Mushroom Substrate + Biochar; T_6 = Spent Mushroom Substrate + Poultry manure; T_7 = Spent Mushroom Substrate + Vermicompost

Figure 12. Relationship between yield (ton/ha) and percent disease incidence at 65 DAT

DISCUSSION

Brinjal is widely farmed and consumed as a vegetable by people all over the world due to its distinct flavor, nutritional and therapeutic value. Iron, calcium, potassium, magnesium, nutritional fiber, protein, and antioxidants are just a few of the vital vitamins, minerals, and nutrients that are included in brinjal. Additionally, it has some phytochemicals with scavenging abilities. Brinjals are rich in anthocyanin chemicals which has strong defenses against diabetes, cancer, and cardiovascular disease. (Naeem and Ugur, 2019). Fusarium wilt caused by *Fusarium oxysporum* f. sp. *melongenae* is a significant issue for brinjal production. Under favorable conditions, it might result in yield losses of up to 70%. (Ashfaq *et al*., 2014). Symptoms of Fusarium wilt include yellowing of leaves, drooping apical shoots which causes death of the brinjal plant. Infected xylem vessel tissue show brown discoloration (Rani *et al.,* 2008.).This soil born pathogern which colonizes the senescing tissues of the infected plant and can live many years in the soil. (Joseph *et al*., 2008). So it is difficult to manage in the field condition. Using resistant variety and use of chemical fungicide can be effective to manage this disease. Russo and Howard (2002) suggested to avoid the use of fungicide on-soil due to its toxic effects.

The current study tested eight different treatments to see how effective they were at controlling Fusarium wilt of eggplant in the field condition produced by *Fusarium oxysporum* f. sp. *melongenae* . Number of infected plants, wilt incidence, plant growth parameters and fruit yield were used to evaluate the effectiveness of the treatments. On the basis of observable symptoms of fusarium wilt of brinjal in response to the selected different treatments were noted at 25, 45 and 65 DAT. All the selected treatments reduced number of infected plants and the disease incidence over untreated control. At 25 DAT the highest no. of infected plants and disease incidence (44.4%) was observed in untreated control and the lowest no of infected plants and disease incidence (5.55%) was observed where spent mushroom substrate + biochar was used. The second lowest disease Incidence (11.11%) was observed in T_6 which was produced by adding poultry manure and spent mushroom substrate. At 45 DAT the highest number of infected plants and disease incidence (55.55%) of fusarium wilt of brinjal was observed in untreated control (T_0) and the lowest number of infected plants

and lowest disease incidence (16.66%) was observed where spent mushroom substrate + biochar was used (T_5) and at 65 DAT the highest disease incidence (77.77%) was observed in untreated

control and the lowest disease incidence (16.66%) was observed in T_5 (spent mushroom substrate + biochar). The second lowest disease incidence was observed in T_6 which was statistically similar to T_1.

In the field study, this is a clear outcome that the treatment T_5 which was produced by adding spent mushroom substrate and biochar effectively controlled the disease incidence in all counting i.e., 25 DAT, 45 DAT, 65 DAT followed by T_6 which was produced by adding spent mushroom substrate and poultry manure. This outcome which was obtained by adding spent mushroom substrate and biochar is partially supported by Ocimati *et al*., (2021), Yusidah and Istifadah, (2018), Salim *et al*., (2017); Adedeji and Aduramigba, (2016). The authors pointed out that spent mushroom substrates have antifungal properties that are effective against *F. oxysporum* because they include a variety of microorganisms that are hostile to pathogenic fungi. This outcome is also partially supported by De medeiros *et al*., (2021), Podeva *et al*., (2021) they described biochar has the capability to reduce the mycelial growth of *Fusarium* spp. either directly through antagonism or indirectly through the induction of systemic resistance in plant. Singh and Kumar, (2020) reported that by decreasing the the growth of chlamydospore biochar decreased upto 50% *Fusarium* spp. growth as compared to the control where the pathogen is growing at a rate of 93%. This result also supported by Wang *et al*., (2019) and Akhter *et al.,* (2016), the authors reported that the application of SMS or SMC as a soil organic amendment along with biochar significantly reduced *Fusarium oxysporum* populations in tomato plants as compared to the control group. The researchers found that the SMS-biochar mixture increased soil fertility and microbial activity, creating a healthier soil environment that inhibited *Fusarium oxysporum* growth and development. Spent mushroom substrate and poultry manure also combinedly reduce wilt incidence in field condition. Similar outputs were obtained by Zeng *et al*., (2022) and Melero-vara *et al*., (2011) the authors reported that poultry manure and other N-rich organic amendments i.e., SMS have the ability to suppress the viability of certain soil-borne plant diseases. The treatments that were used to treat brinjal wilt significantly improved all the evaluated plant growth parameters such as plant height, no. of leaves/plant, no. of branches/plant. The highest result was found in T_5 and the second highest result was found in T_6. The lowest result was found in untreated control T_0. The effect of different treatments on yield and yield contributing characters was also studied and significant variation was observed. In case of fruits per plant, Number of fruits/Plot, individual fruit weight, Fruit length all the treatments effect was found effective, and the highest result was found in T_5 which was statistically similar with T_6 (Spent Mushroom Substrate + Poultry manure).

In case of yield of brinjal, yield/plant, yield/plot, yield (ton/ha) was measured. The performance of the treatments showed promising results as compared to the control. The highest yield (865.0 g/plant, 5.19kg/plot and 12.71 ton/ha) was found in T_5 which was produced by adding spent mushroom substrate and biochar. The second highest result was observed in T_6. The lowest yield (277.67 g/plant, 1.6667 kg/plot and 4.080 ton/ha) was observed in untreated control $T_{0..}$ The present study showed the capability of the treatment to reduce Fusarium wilt incidence and to enhance plant growth parameters (Plant height, No of leaves/plant, No of branches/plant), yield and yield contributing characters (fruits per plant, Number of fruits/plot, individual fruit weight, Fruit length).so it was observed that the treatment T_5 showed tremendous performance in each and every case of yield and yield contributing characters followed by T_6. These findings are partially supported by Nguyen *et al*. (2021). In this study they use the combination of 45% spent mushroom substrate, 30% manure, 7.7% rice husks, 1.5% phosphorus, 0.2% commercial Trichoderma (Tribac), 0.1% rice bran, 15% sand, and 0.5% micronutrients and water as treatment to evaluate the growth, quality, and yield of three muskmelon varieties.These findings also partially supported by Rahman *et al*. (2016). They use Chemical fertilizer + Cowdung (5 t/ha) + SMS (5 t/ha) on the growth, yield and proximate composition of brinjal.

The growth performance and yield were positively influenced by the application of spent mushroom substrate and biochar. The literature in favour of this treatment against Fusarium wilt (*Fusarium oxysporum*) is available in the previous report (Obermeier 2021; Siric *et al.,* 2022). Depending on their composition it might be beneficial to combine SMSs with other organic or inorganic amendments to improve their physical, chemical, and biological properties and thus their capability for enhancing crop yield.

The growth performance and yield of brinjal also increased by using the treatment T_6 (Spent Mushroom Substrate + Poultry manure). This result of the study is partially supported by Rahman *et al.,* (2018). They use combinations of poultry manure and chemical fertilizer as treatment to increase the yield of rice. This outcome goes in the same line which is supported by Frąc *et al*., (2021). The authors reported that application of SMS and poultry manure increased the diversity of fungi, including Tremellomycetes and Pezizomycetes for the SMS additive, while the levels of Mortierellomycetes, Pezizomycetes, and Leotiomycetes increased after the addition of poultry manure. This finding should be helpful in the task of managing the soil mycobiome as well as crop protection and productivity.

SUMMARY AND CONCLUSION

The effect of the treatments for management of Fusarium wilt of brinjal were determined by recording data in terms of number of infected plants, wilt disease incidence (%), yield and yield contributing characters against wilt disease.

All the treatments significantly reduced Fusarium wilt incidence compared to T_0 (control) that ranged from 16.66% to 77.77%. The minimum disease incidence (16.66%) was observed from T_5 which is application of Spent Mushroom Substrate + Biochar .On the other hand the highest disease incidence (77.77%) of fusarium wilt was recorded in control .The second lowest disease incidence was observed by the application of T_6 (Spent Mushroom Substrate + Poultry manure) which was statistically identical with T_1 (Spent Mushroom Substrate). The application of T_2, T_3, T_4, T_7 had moderate effects against wilt disease.

The effects of different treatments were differed significantly in respect of plant growth characters. The highest performance was found in case of the application of T_5 (Spent Mushroom Substrate + Biochar). Here we found the highest plant height (71.0 cm), number of leaves per plant (111), number of branches per plant (11.33). The lowest result was found in T_0 (control) in case of plant height (50.66cm), number of leaves per plant (73.67), number of branches per plant (5.67).

The effects of different treatments were differed significantly in respect of yield and yield contributing characters. The highest performance was found in case of the application of T_5 (Spent Mushroom Substrate + Biochar). Here we found the highest number of fruits/plant (12.27), Number of fruit/Plot (73.66), Individual weight of fruit (92.51 g), Fruit length (9.11cm). The lowest result was found in T_0 in case of number of fruits/plant (3.77), Number of fruit/Plot (22.66), Individual weight of fruit (66.69gm), Fruit length (7.80cm).

The highest yield (865.0 g/plant, 5.19kg/plot and 12.71 ton/ha) was recorded in case of T_5 (Spent Mushroom Substrate + Biochar). The lowest yield (277.67g/plant, 1.6667 kg/plot and 4.080 ton/ha) was found in T_0 (control).

CONCLUSIONS

The effects of different treatments differed significantly in respect of plant growth characters viz. plant height, number of leaves per plant, number of branches per plant compared to control. The highest performance was found in case of the application of T_5 (Spent Mushroom Substrate + Biochar) which was statistically similar with T_6 (Spent Mushroom Substrate + Poultry manure). The lowest result was found in T_0 (control). The effects of different treatments differed significantly in respect of yield and yield contributing characters viz. Number of fruits/plants, Number of fruit/Plot, Individual weight of fruit, Fruit length.

The application of most of the treatments showed similar trend of results in case of yield and yield contributing characters. The highest performance was found in case of the T_5 which was statistically similar with T_6 (Mushroom Substrate + Poultry manure). The lowest result was found in T_0 (control). In case of disease incidence the highest percentage of disease incidence was found in control T0 and the lowest percentage of disease incidence was found in T5 treatment.

RECOMMENDATION

Considering the overall performance of the treatments that applied in the experiment in controlling Fusarium wilt. The application of T_5 (Spent Mushroom Substrate + Biochar) T_6 (Spent Mushroom Substrate + Poultry manure) could be used as eco-friendly approach and may be adviced to the farmers for profitable production. However, further study need to be carried out for consecutive years to include more options as management practices in different Agro Ecological Zones (AEZs) of the country.

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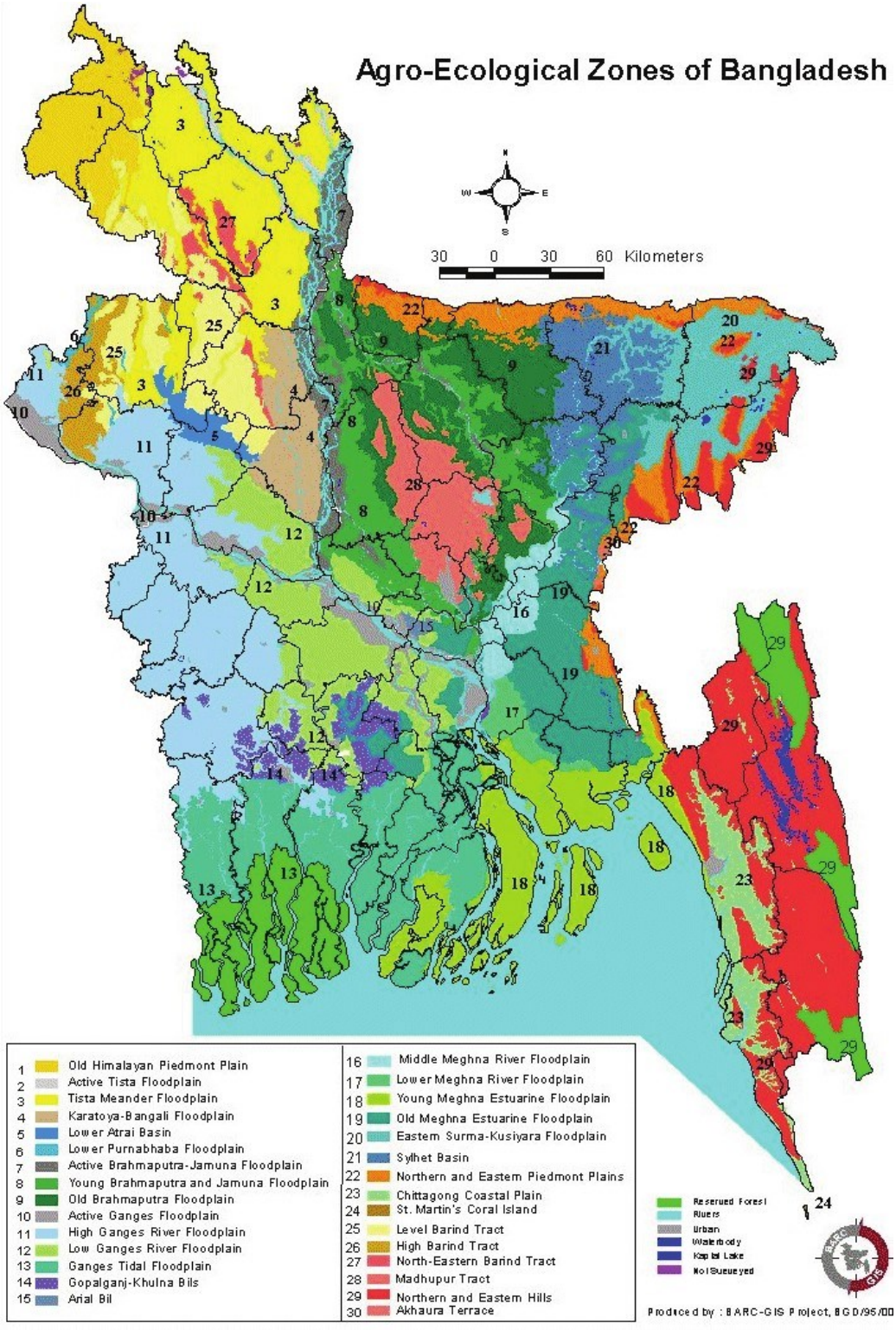
APPENDICES

APENDIX I: AEZ OF BANGLADESH



APENDIX II. MAP OF THE SAU FARM LAND

APPENDIX III. PHYSICAL AND CHEMICAL PROPERTIES OF THE SOIL

CHARACTERISTICS		VALUE
Particle size analysis (%)	% Sand	30
	% Silt	40
	% Clay	30
Consistency		Granular and friable when dry
Soil Textural		Loam to Clay loam
pH		5.6
Bulk Density (g/cc)		1.45
Particle Density (g/cc)		2.53
Organic carbon (%		0.45
Organic matter (%)		0.78
Total N (%)		0.06
Available P (ppm)		20.0
Exchangeable K (meq/100g soil)		0.12

Source: SRDI

APENDIX IV: NUTRITIVE COMPONENTS IN 100 gm OF EDIBLE PORTION OF BRINJAL

Components	Composition
Calories	24.0
Moisture (%)	92.7
Carbohydrates (g)	4.0
Protein (g)	1.4
Fat (g)	1.3
Calcium (mg)	18.0
Magnesium (mg)	47.0
Sulphar (mg)	44.0
Oxalic acid (mg)	18.0
Iron (mg)	0.9
Sodium (mg)	3.01
Copper (mg)	0.1
Chlorine (mg)	52.2
Potassium (mg)	2.0

(source: Internet)

APPENDIX 5. COMPOSITION OF POTATO DEXTROSE AGAR (PDA)

Components	Composition
Potato (Peeled and sliced)	200
Dextrose	20
Agar	20
Water	1000

APPENDIX-6. LAYOUT OF THE EXPERIMENTAL FIELD (RCBD)

T_0 = (Control)

T_1 = Spent mushroom substrate

T_2 = Vermicompost

T_3 = Poultry manure

T_4 = Biochar

T_5 = Spent mushroom substrate + Biochar

T_6 = Spent mushroom substrate + Poultry manure

T_7 = Spent mushroom substrate + Vermicompost