MANAGEMENT OF ANTHRACNOSE OF CHILLI THROUGH SELECTED FUNGICIDES AND PLANT EXTRACTS

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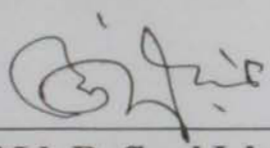

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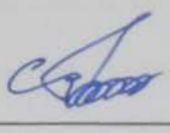

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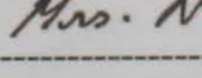

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This is to certify that the thesis entitled, "**MANAGEMENT OF ANTHRACNOSE OF CHILLI THROUGH SELECTED FUNGICIDES AND PLANT EXTRACTS**" submitted to the Department of Plant Pathology, Faculty of Agriculture, Sher-e-Bangla Agricultural University, Dhaka, in partial fulfillment of the requirements for the degree of *MASTER OF SCIENCE IN PLANT PATHOLOGY*, embodies the result of a piece of bona fide research work carried out by *MUHAMMAD MAZHARUL ISLAM, Registration No. 27565/00727*, under my supervision and guidance. No part of the thesis has been submitted for any degree or diploma in any institutes.

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

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TO MY

BELOVED PARENTS

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CONTENTS

MATERIALS AND MATHODES CONTD.

LIST OF TABLES

LIST OF FIGURES

LIST OF APPENDICES

MANAGEMENT OF ANTHRACNOSE OF CHILLI THROUGH SELECTED FUNGICIDES AND PLANT EXTRACTS

ABSTRACT

The effect of twelve treatments *viz.* Bavistin 50 WP (T_1), Cupravit 50 WP (T_2), Dithane M-45 (T_3), Proud 250 EC (T_4), Ridomil Gold (T_5), Tilt 250 EC (T_6), Allamanda leaf extract (T_7), Neem leaf extract (T_8), Garlic extract (T_9), Ginger extract (T_{10}), Onion extract (T_{11}) and Control (T_{12}) were tested against *Colletotrichum capsici* causing Anthracnose of chilli during winter season (2006-2007) at the Farm of Sher-e-Bangla Agricultural University, Dhaka. The highest seed germination (80%) was obtained from ginger extract and neem extract treated seeds in laboratory condition while the germination was 78.83% and 74.00%, respectively ginger and neem extract in the field condition. The efficacy of the treatments varied significantly in terms of disease incidence, disease severity and yield of chilli. Tilt 250 EC (T_6), Proud 250 EC (T_4), Cupravit 50 WP (T_2) and Allamanda extract (T_9) showed promising effect against the disease. Application of Tilt 250 EC increased 195.90% fruit yield of chilli followed by Proud 250 EC (177.59%) and Bavistin 50 WP (159.56%) against anthracnose disease of chilli. Cost benefit analysis showed that the application of Tilt 250 EC (T_6) resulted the highest BCR (4.07) followed by Proud 250 EC (3.68), Bavistin 50 WP (3.47), Allamanda leaf extract (3.16) and Dithane M- 45 (2.95) compared to control.

CHAPTER ONE

INTRODUCTION

CHAPTER I

INTRODUCTION

Chilli (*Capsicum annuum* L.) belongs to the family Solanaceae is the most important spice crop in Bangladesh. It is cultivated in almost all tropical and sub tropical countries in the world. Chilli is grown in all parts of Bangladesh. Chilli fruits are consumed as fresh, dried or processed product, as table vegetables as well as spices or condiments. It is a rich source of vitamin A, E and C. Chilli contains 1.29 mg protein, 11 mg calcium, 870 IU, 17.5 mg ascorbic acid, 0.06 mg thiamin, 0.03 mg riboflavin and 0.55 mg niacin per 100 mg edible fruit (Joshi and Singh, 1975). It has an important role in both winter and summer to fulfill the spice demand of our country. Chilli is grown in all season and all areas of Bangladesh .The total cultivation area is 65000 hectare where 23417 ha in Kharif season and 41583 ha in Rabi season with total production of 52000 MT and annually average yield is 0.80 MT in 2004-2005 (Anon., 2007). Of many reasons for high price of chilli, lower production rate is an important factor.

Incidence of insect pests and diseases greatly hamper the production of chilli. This crop suffers from the various diseases; about 83 different diseases, of which more than 40 diseases are caused by fungus (Anonymous, 1960 and Rangaswami, 1979). Among the fungal diseases, at least 31 diseases are transmitted through seeds (Richardson 1983, Deena and Chowdhury 1984 and Sharma 1985). Among the seed borne fungal diseases, anthracnose is one of the major and devastating disease of chilli in the country. It is caused by *Colletotrichum capsici* (syd). In Bangladesh, the disease is common in flooded low lands where Solanaceous spices are grown continuously without crop rotation.

Management strategies for this disease include use of healthy seeds and transplants, resistant cultivars and fungicidal sprays. At present, plant diseases are mainly managed by the use of chemical fungicides. The indiscriminate use of chemicals is not only hazardous to living being but also breaks the natural ecological balance by killing the antagonist microorganisms. Now a day's induced resistance in plants against the pathogen is drawing the attention of plant pathologists for successful management of crop diseases. But the resistant variety of chilli against anthracnose diseases is not available. Among the control measures, seed treatment is probably the cheapest and safest method to control anthracnose pathogen. It is reported that Mancozeb and Carboxin completely controlled the seed- borne *Alternaria alternata*, *Fusarium moniliforme and Colletotrichum capsici* (Mridha and Chowdhury 1990).

Botanical control of seed-borne pathogens of many crops tested in many laboratories and field trials showed that crude extract from rhizome, leaf, stolen fruits and cloves of garlic (*Allium sativum),* marigold *(Tagetes erecta),* neem *(Azadirachta indica),* dholkalmi *(Ipomoea fistulos.L.)* and ginger *(Zingiber officinale)* could restrict growth of the seed-borne fungi. It was found that garlic extract at different concentration significantly reduces seed borne *Colletotrichum corchori*, *Fusarium spp* and *Macrophomina phaseolina* in jute (Khan and Fakir, 1995). Neem extract minimize the ripe fruit rot of chilli. Garlic extract can effectively control chilli anthracnose (Harbant *et al.*,1999). *Colletotrichum capsici* was effectively controlled by the leaf extract of *Solanum torvum, Dutura metel* and *Prosopis juliflora* (Gomathi and Kannabirran 2000). Thus, attempts need to be taken to manage the anthracnose of chilli through environment friendly plant extract including fungicides.

Considering the above facts, the present study was under taken to achieve the following objects:

1. To determine the efficacy of selected fungicides and plant extracts against *Colletotrichum capsici* causing anthracnose of chilli.
2. To determine the Benefit Cost Ratio (BCR) of effective fungicides and plant extracts applied against anthracnose of chilli

CHAPTER TWO

REVIEW OF LITERATURE

CHAPTER II

REVIEW OF LITERATURE

2.1 Management through fungicides

Juangbhanich and China (1975) observed that seed treatment with Ceresan (Ethoxyethylmercury) at 0.16% or Desline (Methyl 2- benzimidazole carbamate) at 0.8% of seed wt gave complete control of *Colletrotichum peperatum* and *Colletotrichum capsici.*

Bhuiyan and Fakir (1982) got 90% control of *Colletotrichum dematium* and *Macrophomina phaseolina* when treated the soybean seeds with Captan (0.5%), Homai 80 WP (0.25%), Topsin M 70 WP (0.5%) and Vitavax-200 (0.35%) while all these fungicides contorlled 70 to 95% seedling diseases caused by the two fungi in pot experiment. The found Homai 80 WP and Captan as the best fungicide to control two major seed borne pathogen of soybean.

Ahmed (1986) recommended Delsan (0.2%) and Cuman-L (0.5%) against anthracnose of chili caused by *Colletotrichum capsici*; Bordeaux mixture against anthracnose of betel vine caused by *Colletotrichum capsici*; Dithane M-45 (0.2%) against anthracnose of mango caused by *Colletotrichum gloeosporioides* and Copper oxychloride (0.3%) or Dithane M-45 (0.2%) against anthracnose of guava for the management of the diseases respectively.

Thind and Jhooty (1987) stated that the most effective and economical control of *Colletotrichum capsici* and *Alternaria alternata* on *Capsicum annuum* under Panjab conditions given by sprays with Difolatan (Captafol) at 0.2 %, followed by Dithane M-45 (0.2 %).

Eswaramurthy *et al.* (1988) found that spraying of 10 fungicides twice at 15 days interval, the best control of *Colletotrichum capsici* on *Capsicum annuum* was given by Foltaf (captafol) at 0.2%, followed by Fytolan (Copper Oxychloride) at 0.25% and Bavistin (Carbendazim) at 0.1%. These compounds also gave good control of the die-back phase of the disease and increased yields.

Perene and Joi (1989) found that the treatment of chilli seeds with Thiram or Bavistin and fungicidal spray of seedling with Dithane M-45, Blitox, Carbendazim were more effective than either of the single treatment in controlling *Colletotrichum capsici* . The incidence of the disease was least after seed treatment with Bavistin followed by spraying with Dithane M-45. Fruit rot was less with combination effect of seed treatment with Thirum and spraying with Mencozeb.

While working with anthracnose disease of guava caused by *Colletrotichum gloeosporiodes,* Hossain (1989) observed that Topsin M, Rovral 50 WP, and Rovral Flo completely inhibited the growth of the fungus. When applied on plant Topsin M significantly reduced fruit infection and disease severity followed by Rovral Flo, Rovral 50 WP and Dithane M-45. The author concluded that when the number of fungicidal sprays increased from 4 to 7 the fruit infection were reduced and fungicidal efficacy was significantly improved.

Datar *et al.* (1990) conducted experiment on fungicidal control of anthracnose of chillies and found that in field trials over three consecutive seasons, (1984-87) three sprays of mancozeb 0.25 %, fortnightly from one month after transplanting gave the best control of *Collectotrichum capsici* on the susceptible *Capsicum annuum* cv. Jwala, with highest yields and lowest incidence of fruit rot.

Raj *et al.* (1990) stated seed treatment with thiram + carbendazim followed by 3 sprays of carbendazim gave minimum disease incidence of anthracnose of bean (*Colletotrichum capsici*) and maximum yield per plot. Seed treatment and 3 sprays of mancozeb also increased yield and reduced disease incidence. Srivastava and Soni (1993) reported that 0.1% Bavistin and 0.25% Dithane M-45 (Mancozeb) were effective fungicides for the control of the disease under laboratory and field conditions.

Sinha (1990) observed on the field trials during 1984-87, with seven commonly available fungicides. The best control of *Colletotrichum capsici* on *Capsicum annuum* was given by using Folaf (Captafol). The best cost-benefit ratio was obtained with Dithane M-45 (Mencozeb) and Bilotox 50 (Copper oxychloride).

Biswas (1992) found in the field trials of six fungicides, Bavistin, Mac sulpher, Dithane M-45, Knowin, Mancozeb and Thiram against *Colletotrichum capsici* on *Capsicum annuum* the best control was given by Bavistin 50 WP at 0.1% concentrations.

Rahman *et al.* (1994) evaluated Tilt 250 EC, Pencozeb 80 WP, Topsin-M 70 WP, Knowin 50 WP and fungi-kill 50 WP against *colletotrichum lindemuthianum* causing anthracnose of country bean. Among the fungicide tested Knowin 50WP was found to be the best followed by Tilt 250 EC and Topsin 70 MP both *in vitro* and *in vivo* controlled of the *Collectotrichum lindemuthianum.* Tilt 250 EC was found to be the best performance followed by Knowin 50 WP both *in vitro* and *in vivo* condition in controlling *Colletotrichum dematium.*

Acharya and Das (1995) conducted a field experiment for the control of *Collectotrichum capsici* causing anthracnose of Piper betel. Foliar spray of Bitertanol (0.05 %), Mancozeb (0.2%) Ziram (0.1%) and Bordeaux mixture (0.5%) were found to be effective against the fungus. A higher cost-benefit ratio was observed for Mancozeb and Bordeaux mixture.

Ebenezar and Alice (1996) conducted field experiments to study the efficacy of 9 commonly used fungicides in controlling fruit rot and dieback of chilli caused by *Collectotrichum capsici* in Tamil Nadu, India. The best control was achieved with Mancozeb (0.2%) followed by Carbendazim (0.2%) and Copper oxychloride (0.2%).

Kumawat (1997) conducted a field experiment at Jalore, India during the Kharif season, to evaluate eight fungicides against anthracnose disease of chilli. Three sprays of fungicides Mancozeb, Thiophanate- methyl, Catafol, Ziram and Carbendazim at 15 days intervals significantly reduced the disease intensity.

Haque *et al.* (1998) tested five fungicides against seed borne fungi of chilli and reported that Vitavax-200 totally eliminated the seed borne infections of *Colletotrichum capsici* and increased germination.

Romer *et al.* (2002) conducted a series of field trial in Germany and in the Azores in 1998. Seed treatment and fungicidal spray had been tested for their efficiency to control anthracnose. They reported that the most efficient fungicides were "Harvesan" (Carbendazim + flusilazole) + "Amistar" (Azoxystrobin) (Germany + Azores), "Simbo" (fenpropimorph + propiconazole) (Germany), "Alto 100 SL" + "Bravo 500" (Cyproconazole + Chorothalonil) (Germany + Azores), Folicur + Bravo 500 (Tebuconazole + Cholothalonil) (Germany), "Amistar" (Germany) and "Harvesan" (Germany + Azores).

Saleem (2000) conducted a survey during 1996-1999 and more than 150 diseased *Piper beetle* specimens were collected in Thatta and Pakistan. *Colletotrichum capsici* and *Colletotrichum gloeosporides* were isolated and identified. Benoyl and Thiophanate-methyl gave good control of these pathogens.

Hedge and Anahosur (2001a) also conducted a field experiment to determine the effective fungicidal treatment against fruit rot disease of chilli (*Capsicum annuum*) under rainfed conditions. The efficacy of non-systemic (Mencozeb, chlorothalofil, Copper oxychloride, and Iprodione at 0.3% concentration) and systemic fungicides (Cardendazim, Tridomorph, and Hexaconazole at 0.15% concentration) and botanical fungicide Nimbicidin at 3% were tested. Among the tested fungicide, Cardendazim showed the least percent disease index (30.96%), followed by Propicanazole (36.53%) and Hexaconazole (35.63%). Iprodione yielded

the highest percentage disease index (71.57%). Cardendazim was the best fungicide for controlling fruit rot disease. The fungicides also helped in obtaining maximum capsicin, ascorbic acid and total sugar contents in chilli fruits.

Hedge and Anahosur (2001b) conducted an experiment on the control of chilli fruit caused by *Colletotrichum capsic.i* The fruits were treated with 10%, 25%, 50% respectively and saturated solution of sodium chloride for 10,15 and 30 minutes, stored in different storage conditions. They observed all treatments differed significantly regarding the to disease incidence. Treatment with sodium chloride at 50% concentration or saturated solution for 30 minutes reduced the disease in storage to the maximum extent. Storing chilli fruits after treatment with either 50% or saturated salt solution for 30 minutes under sunshine was the best from other in getting healthy dried fruit.

Deeksha *et al.* (2002) found that chemical control of anthracnose of *Vigna mungo* where seed treatment followed by two prophylactic sprays of Bavistin (Carbendazim) or Tilt (Propiconazole) at 0.1% concentration at 15 days interval showed minimum disease severity and maximum grain yield followed by spraying 1% Contaf and 2% Indofil M-45 in the plots.

Deshmukh *et al.* (2002) conducted an experiment for comparison between Mancozeb at 0.25% and biological pesticide Zetron at 0.2, 0.25 and 0.4% in controlling anthracnose of chilli. The Zetron significantly reduced the growth of the fungus comparison to the control. The development of lesions on chilli fruits was considerably slower with the lowest lesions development resulting from the Mancozeb treatment followed by Zetron at 0.4, 0.25 and 0.2 %.

Ekbote (2002) conducted an experiment against fruit rot of chilli to evaluate the efficacy of Copper hydroxide at 0.10, 0.15, 0.20 and 0.25%; Chlorothalonil at 0.20%; and Carbendazim at 0.10%. The lowest disease index (30.47%) resulted from the application of 0.10% Carbendazim (30.47) followed by copper hydroxide (37.70%) sprayed at 0.25 % concentration. Crop yield was the highest with application of Copper hydroxide (0.25 %). The results indicated the potential of using Copper hydroxide in controlling *Colletotrichum capsici* to avoid the development of the resistance of the pathogen to carbendazim.

Rajabaskar *et al.* (2002) conducted three experiments to investigate the harvest time residues of Tilt 250 EC (Propiconazole) in local chilli cultivar. Propiconazole was applied at 125 and 250g a.i./ha, at 15 days interval; starting from 45 days after transplanting. The residues on the crop were analyzed from the sample collected 15 days and 30 days after application. The residues were extracted with methanol, cleared using dichloromethane (Methyl choloride) and analyzed by GC-ECD. The harvest time resudes of Propiconazole applied at recommended rate of 125 g a.i./ha were below the detectable limit 15 and 30 days after application while propiconazole applied at 250 g a.i./ha resulted in residue concentration of 0.074, 0.098 and 0.167 ppm.

Ekbote (2003) conducted a field experiment during the 2000/01 and 2001/02 kharif seasons in Karnataka, India to evaluate the efficacy of Prochoraz compared with Carbendazim and Mencozeb against fruit rot and die-back (*Colletotrichum capsici*) of chilli. The treatments comprised of 0.05, 0.75, 0.1 and .0125% Prochoraz 45 EC; 0.25% Mencozeb; 0.25% Copper oxychloride 75 WP and .01% Carbendazim. Among these

fungicides 0.125% Prochoraz gave the lowest PDI for die back and fruit rot and highest yield performance.

Mandal and Beura (2003) conducted an experiment on chilli cv. Utkal Ava at Orissa, India to determine the efficacy of different application dates of 0.05% Mono crotophos + 0.25% Mancozeb against chilli thrips (*Scirtothrips dorsalis*), anthracnose and ripe fruit rot (*Colletotrichum capsici*). The pesticide combination was applied at 25, 45 and 65 days after transplant (DAT); at 25 and 45 at 25 and 65, and 45 and 65 DAT; and at 25, 45 and 65 DAT. Leaf curling rate, percent disease index (PDI) of anthracnose and PDI of ripe fruit rot were reduced.

Joi *et al.* (2004) conducted a field trial during kharif 2002/03, Maharastra in India to evaluate the efficacy of RIL 006/CI 0.05, 0.1, 0.15 and 0.2% concentration against anthracnose of chilli caused by *Colletotrichum capsici.* They also used Shan at 0.1%, Contaf (Hexaconazole) at 0.3% and 0.1%. They observed all treatments except for RIL 006/C1 at 0.05% were significantly superior over the untreated control in reducing intensity of leaf anthracnose and fruit rot infection. RIL 006/C1 at .02%, Tata Shan at 0.1% + Contaf at 0.3% and RIL 006/C1 at 0.1% reduced anthracnose and fruit infection 70.7, 67.1 and 62.6%, respectively over control.

Rahman *et al.* (2004) conducted on experiment in Mymensingh, Bangladesh to evaluate the efficacy of Bion (0.005%), Azoxystrobin and Carboxin in inducing systemic resistance to Anthracnose (*Colletotrichum Capsici*) in chili (*Capsicum annuum*). They observed die-back symptom did not appear in Bion treated seeds but was recorded in Azoxystrobin and Carboxin treated seeds. Lesion size, leaf infection and leaf area damage were less in plants grown from Bion and Azoxystrobin treated seeds. Bion

treatment resulted in moderate resistance to anthracnose, where Azoxystrobin and Carboxin treatment resulted in susceptibility of the crop to the disease.

Anon. *(1985-2005)* conducted an experiment to evaluate the fungicides namely, Indofil, Dithane M-45, Topsin M and Tilt 250 EC for controlling the anthracnose of chilli. Data indicated that incidence of anthracnose significantly lower in Topsin M treated plot which was significantly similar to Indofil sprayed plot. Tilt 250 EC and Dithane M-45 were also effective.

Roy (2005) sprayed eleven fungicides for four times at ten days to control *Colletotrichum capsici* on chilli (*Capsicum annuum*). Fild 250 EC at 0.05%, give the effective result followed by Propicon 250 EC, Copper 50 WP and Indofil Z-78. These compounds also could control of the die back phase as well as anthracnose disease and increased yield of chilli.

2.2 Management through plant extract and phytoncydes

Misra and Dexit (1977) reported the fungitoxicity of garlic bulb and they showed that they showed that garlic extract could be used be as a potent fungicide against plant pathogens *in vitro.*

Ahmed and Sultana (1984) used bulb extract of garlic at different concentration that inhabited the spore germination and mycelial growth of some important fungal pathogens of jute such as *Macrophomina phaseolina, Botryodiplodia theobrome* and *Colletotrichum corchori.* Jute seeds treated with garlic paste had increased rate of germination and decreased rate of post emergence seedling mortality.

Mali and Joi (1985) tested seven fungicides against seed borne mycoflora of chilli and reported that Vitavax-200 (carbendazim) was the most effective against colony growth and sporulation *Colletotrichum capsici.*

Assadi and Behroozin (1987) found that garlic extract was more active than onion inhibiting the growth of *Fusarium solani*, *Fusarium oxysporum* and *Fusarium acuminate.*

Kasem and Vijai (1987) tested the effects of some medicinal plants on growth of fungi and found potential in plant disease control. Ten medicinal plants, Stemona, nux-vomica tree, derris, urging croton, star anise, clove tress, garlic and care way were tested for this antifungal property to some fungal species namely–*Phytophthora spp.*, *Phythium aphanidermatum, Rhizopus microsporus, Alternaria alternata* and *Fusarium solani.* Staranise at the concentration of 2000 ppm completely inhibited growth of all test fungi followed by caraway, lemon grass, clove tree and garlic, respectively

Lakshmonsn *et al.* (1990) found that garlic clove extract was most effective in inhibiting mycelial growth and spore germination of *Corynespora cassiicola.*

According to Tariq and Magee (1990) volatile components of crude aqueous extracts of garlic bulb was effective in inhibiting germination of micro conidia and reduction of hyphal extention of *F. oxysporum f. lycopersici* in anemic culture.

Dubey and Dwivedi (1991) observed fungitoxic properties of extracts of leaves and bulb of onion, garlic and fruit and bark extract of *Allium*

cearavica against vegetative growth and sclerotial viability of *Macrophomina phasttolina.* They also found that all the extracts inhibited growth of the fungus while garlic bulb extract was more effective than other extracts employed in the tests.

Achimu and Schloesscr (1992) studied the effect of neem seed extracts against downy mildew *(Plasmopara viticola)* of grapevine. They found that raw neem seed extract and commercial neem products (margo-san-O, neem oil and neem- Azal-S) had high (80-90%) antifungal properties against *P. viticola.* They concluded that the antifungal property of neem products could be attributed to an inhibition of the indirect germination of sporangia.

Fakir and Khan (1992) reported that garlic bulb extract al different concentrations reduced the seed borne infection in jute. Both concentrated garlic extract and Vitavax-200 were more or less equally effective in controlling *M. phaseollina* reducing 90.9% and 87.9% seed borne infection of the pathogen, respectively.

Hossain and Schlosser (1993) found neem extract to be effective against *Bipolaris snrokiniana.*

Arun *et al.* (1995) found that the extract of garlic bulb was effective in suppressing radial growth of *Fusarium sp* and *Colletotrichum capsici*

Mohanty *et al.* (1995) investigated the allelopathic control of *Phomopsis vexans,* causal agent of Phomopsis fruit rot of brinjal by aqueous leaf extracts of five plants. Fungal growth was inhibited to a maximum by leaf extract of *Allamanda cathertica* (93.75%) followed by *Aegle mermelos* (85.38%). Leaf

extracts of *Catheranthus roseus*, *Polyalthia longiflora* and *Azadirachta indica* were equally effective, but that of *Ocimum sanctum* was the least effective causing 52.23% growth inhibition.

Kuprashvile (1996) used extracts of garlic bulb for seed treatment of eggplant infected by *Phomopsis vexans* .The results showed that the plant extracts disinfected seeds and increased yields.

Panda *et al.*, (1996) tested the efficacy of leaf extracts from *Polyalthia longiflora, Aegle mermelos, Azadirachta indica, Catheranthus roseus, Ocimum sanctum* and *Allamanda cathertica* for control of Phomopsis blight (caused by *Phomopsis vexans*). Leaf extracts of *Allamanda cathertica* had excellent potential as a fungicide.

Kurucheve and Radmavathi (1997) assayed five selected plant products for fungi toxicity against *Pythium aphanidermatum* (Edson) Fitz, the casual organism of damping off of chillis, Among them extract from *Allium sativum* (garlic) bulb (10%) recorded the minimum mycelium growth (176.00 mg) followed by *Lawsomia incemis* leaf extract. Maximum percentage of seed germination, growth and vigor of chilli seedlings were also observed with garlic bulbs.

Singh *et al.* (1997) evaluated plant extracts of *Catharanthus roseus*, *Azadirachta indica, Pongamia pinnata*, *Tagetes erecta* and onion bulb extracts and garlic extract at 1, 2, 3 and 4% concentrations for radial growth and spore germination of *Colletotrichum capsici*, causing of dieback of Chilli (*Capsicum sp.*). Extracts of garlic bulb at 3% concentration completely inhibited the growth and spore germination of *C. capsici* whereas, 4% extracts of onion bulbs, *A. indica* leaf, *P. glabra* leaf

and *T. erecta* leaf gave complete inhibition of fungal mycelial growth and spore germination.

Khan *et al.* (1998) applied four neem based products, namely Nemokil, Nemokil-S, SDS and SDC and found anti fungal activity against the guava wilt and the anthracnose pathogen. These neem based products however, proved less effective against *Colletotrichum gloeosporioides*.

Moniruzzaman and Ashrafuzzaman (1998) applied garlic (*Allium sutivum*), neem (*Azadirachta indica*) and tobacco (*Nicotiana tabacum*) extracts against Alternaria blight of mustard- Crude extracts of these three plants were significantly reduced disease incidence and severity. In promoting plant height number of siliqua per plant, number of seeds per siliqua, thousand seed weight and yield per plant were observed and the garlic bulb extracts (1:1) was the best among the treatments.

Harbant *et al.* (1999) evaluated some plant extracts for the control of *Colletotrichum capsici* (SYD). The efficacy of neem (*Azadirachta indica*), garlic (*Allium sativum*) and Tagak-tagak (*Rhinocanthus indica*) at 5000 ppm on *Capsicum anmuum* was compared with the fungicide carbendazim (Bavistin) at 100 ppm. Garlic extract performed well under room humidity, while Tagak-tagak extract showed good control of Chili anthracnose under high moisture conditions. Neem extract minimized the ripe fruit rot of chilli.

Khan (1999) studied the effect of plant extracts (Allamanda, Bel and Neem) for the management of Phomopsis blight/fruit rot of eggplant in field condition. Among the three plant extracts, Allamanda was the most effective.

Howlader (2003) observed that seed treatment with allamanda leaf extract (1:1) effectively increased germination of eggplant seeds and tremendously decreased nursery diseases.

Meah (2003) reported that garlic bulbs extract (1:10) and allamanda leaves extract efficiently controlled *Phomopsis vexans* in the laboratory, nursery house and in the field reducing severity of leaf blight and fruit rot by 71-75%.

Kumaran *et al.* (2003) evaluated chemical fungicide Mancozeb at 320 ppm and ethanolic extract of the roots of 18 different plant species for their fungi toxic activity against anthracnose of chilli. They observed ethanolic root extract of *Abrus precatorious* and *Rauvolifia tetraphylla* showed significant inhibitory effects on both the conidial germination and radial growth of *Colletotrichum capsici.*

Islam (2004a) found garlic bulbs and allamanda leaves extract caused 76-100% inhibition of mycelial growth of *Phomopsis vexans*. Diethyl ether, Dichloromethane and water acted as effective solvents for spraying garlic extract. TLC studies showed the presence of a number of compounds having very low to high polarity in garlic bulbs and allamanda leaves extracts.

Yesmin (2004) reported that Neem leaf extract was most effective in controlling canker of leaf and fruit caused by *Xanthomonas citri*. Garlic extract and Neem leaf extract were the most effective in controlling die back of twig and branch of citrus respectively. She also recommended that citrus canker can successfully controlled by Neem leaf extract and garlic extract.

CHAPTER THREE

MATERIALS AND METHODS

CHAPTER III

MATERIALS AND METHODS

A field experiment was conducted in the farm of Sher-e-Bangla Agricultural University, Dhaka, Bangladesh during the period of November'06 to May'07 for the management of anthracnose of chilli through some fungicides and plant extracts. The detail materials and methods of this experiment are presented in this chapter-

3.1 Experimental site

The experiment was conducted in the Plant Health Laboratory of the Department of Plant Pathology of Sher-e-Bangla Agricultural University and the farm of the Sher-e-Bangla Agricultural University, Dhaka-1207 (Fig. 5). The Site of experimental plot lies in the 23° 74 N latitude and 90 ° 35 E longitudes with an elevation of 8.2 meter sea level (Anon. 1989).

3.2 Characteristics of soil

The soil of the experimental area was non calcareous dark grey and belongs to the Modhupur Tract (UNDP, 1988) under AEZ 28. The selected plot was medium high land and the soil series was Tegaon (FAO, 1988). The p^H of the soil was 5.8. The characteristics of soil under the experimental plot were analyzed in the SRDI, soil testing Laboratory, Khamarabari, Dhaka and the details of the soil characteristics are presented in Appendix 1.

3.3 Weather condition of the experimental site

The geological situation of the experimental site was under the subtropical climate, characterized by three distinct seasons, the monsoon or rainy season from November to February and the pre- monsoon period or hot season from March to April and monsoon period from May to October (Edris *et al,* .1979). Average monthly maximum and minimum temperature were 25.3°C and 11.15°C respectively. Details of the metrological data of air temperature, relative humidity, rainfall and sunshine during the period of the experiment was collected from the Bangladesh Meteorological Department (Climatic Division) and presented in Appendix 2.

3.4 Experimental period

The experiment was carried out during Rabi Season in 2006-2007 starting from November '06 to May' 07.

3.5 Cultivar's used in the experiment

Chilli (*Capsicum annuum L.)* variety Bangla Lanka (BARI Morich-1) was used in the experiment. The seed were collected from regional spice center, Bangladesh Agricultural Research Institute (BARI) at Joydebpur Gazipur.

3.6 Seed treatment

Seeds of chilli were subjected to seed treatment. Two hundred seeds for each treatment were soaked or dipped for 12 hours in fresh water. Two hundred seeds of each treatment were dipped into the respective solution separately for seed treatment. Seeds were soaked for five minutes. After soaking, seeds were transferred to a newspaper sheets for drying. Then it was ready for sowing in the seedbed.

3.7 Incubation of seeds for germination percentage

In the lab condition, the effect of different fungicides and plant extracts on seed germination was determined by the blotter method according to International Rules for Seed Health Testing (Anon., 1976). Twenty five seeds were plated on 3 layer moist blotter paper in 9 cm petri dish maintaining uniform distance. The blotters and Petri dishes were sterilized properly before plating. Plating was done on the 29th November 2006. The plates were incubated at 25 ± 4°C temperature for 21 days or three weeks. After incubating, data on the seed germination were recorded. Each treatment was followed by this procedure separately. Data were recorded three times with seven days interval.

In field condition, two hundred seeds were sown in seedbed after seed treatment. First germination or emergence was observed on 11th November 2006 and first data were recorded on the 3rd December 2006. Data recording were repeated four times with seven days intervals. The germination percentage was recorded using the following formula:

$$\%\ \text{Germination} = \frac{\text{No. of germinated seed}}{\text{No.of total seed}} \times 100$$

3.8 Raising of seedlings

Seedlings were raised in Farm's seedbed of Sher-e-Bangla Agricultural University with proper care and management. The seedbed was prepared by mixing of Urea, TSP, MP, Furadan 3G and well decomposed cow dung. The seed was sown in seedbed and covered with soil. Before sowing seeds were treated with different assigned treatment and create worm condition for quick emergence. The seeds were sown on 03 November, 2006. Two hundred seeds were sown for each treatment. Watering was done to maintain the soil moisture. Shade was provided to save the young and delicate seedling from heavy showering and scorching sunlight. (Islam, 2005)

3.9 Design and layout of the experiment

The field experiment laid out in the Randomized Complete Block Design (RCBD) using three replications. The field was divided into three blocks with 12 unit plots in each. The size of the individual unit plot was 2.0 m × 1.5 m. Block to block and plot to plot distance were 1.0 m and 0.5 m respectively. Layout of the experimental field was presented in Appendix - 4.

3.10 Treatments

Twelve different treatments designated by T_1, T_2, T_3, T_4, T_5, T_6, T_7, T_8, T_9, T_{10}, T_{11} and T_{12} were employed in the experiment. The treatments consist of six fungicides and five plant extracts and a control. The treatment were as follows-

T_1 = Bavistin 50WP

T_2 = Cupravit 50WP

T_3 = Dithane M-45

T_4 = Proud 250EC

T_5 = Ridomil Gold

T_6 = Tilt 250 EC

T_7 = Allamanda leaf extract

T_8 = Neem leaf extract

T_9 = Garlic extract

T_{10} = Ginger extract

T_{11} = Onion extract

T_{12} = Control (Untreated)

3.11 Land preparation

The land was prepared by thoroughly ploughing and cross ploughing with a power tiller followed by laddering. The weeds and rubbishes were removed from the field. The bigger clods were broken and leveled properly into fine soil particles for seedling transplanting.

3.12 Application of fertilizer and manures

After opening the land well decomposed cowdung was applied and thoroughly mixed with soil through frequent ploughing. Before final land preparation inorganic fertilizers were applied. Fertilizer and manure were applied on the basis of chilli production manual (Anon., 2005).

Table 1. Fertilizer and manure applied in the field

Fertilizer or manure	Amount/ha	Required amount for 3.0 sqm plot in gram
Urea	210Kg	150g
TSP	300 Kg	350g
MP	200 Kg	75g
Boric acid	05 Kg	2.5g
Gypsum	110 Kg	125g
Zinc Sulphate	15 Kg	5g
Cow dung	10 Ton	300g

1. All cow dung, TSP, Boric acid, Gypsum and Zinc Sulphate and half MP applied during land preparation.
2. Urea and remaining half of MP applied in three installments as top dressing.

3.13 Transplantation and care of seedlings

Healthy and uniform sized seedling of 45 days old were uprooted from the seedbed in the treatment wise by avoiding any injury to the roots and transplanted on 19 December 2006 in main field (Bose and Som, 1990). Twenty seedlings were transplanted in each plot in the afternoon. For easy lifting of the seedlings from the seedbed moistened enough before uprooting. Watering and news paper shading of the seedlings was done in every morning for the first seven days after transplantation. After establishment of the seedlings, the soil of the plot was loosened from time to time with the help of a khurpi. For the proper establishment of the roots and shoots watering was done up to the harvest as and when necessary.

3.14 Intercultural operation

3.14.1 Shading: Transplanted seedlings were covered with news paper cap (hand made) to protect the delicate seedlings from scorching heat of sunlight.

3.14.2 Irrigation: Watering was done immediately after transplanting the seedlings and continued up to 7 days to keep the soil moisture in suitable condition. The field plots were irrigated five times at 15 DAT (Day after transplanting), 40 DAT, 65 DAT, 80 DAT and 110 DAT.

3.14.3 Gap filling: Gap filling was done when any seedling failed to established and died in the field. More than 50 seedlings were re-transplanted to fill the gaps.

3.14.4 Weeding: Weeding was done just before fertilizer application. Weeding was done four times at 25 DAT, 45 DAT, 80 DAT and 100 DAT.

3.14.5 Use of insecticide: Aktara 0.2% was sprayed used @ three times starting from 15 days after transplanting (DAT) followed by flowering stage and 80 DAT to control chilli aphid and whitefly.

3.15 Fungicides used

Six fungicides *viz.* Bavistin 50 WP, Cupravit 50 WP, Dithane M-45, Proud 250 EC, Ridomil Gold, and Tilt 250 EC were used in the experiment. Details of the fungicides used are given next page:

able 2. Particulars of the fungicides used in the experiment

	Common name	Chemical name	Active ingredient	Dose used
T_1	Bavistin 50WP	Methyl-2-Benzamidazole Carbamate	Carbendazim 50%	1 g/litre
T_2	Cupravit 50WP	Copper oxychloride ($CuOCl_2$)	Copper oxychloride 50%	7 g/litre
T_3	Dithane M-45	Manganous ethylene bisdilhio - carbamate	Dithiocarbamate (80%)	4.40 g/litre
T_4	Proud 250 EC	Propiconazole	Propiconazole 25%	1 ml/litre
T_5	Ridomil Gold	Mencozeb+Metalixil	4% Mencozeb +68% Metalixil	2 g/litre
T_6	Tilt 250 EC	Propiconazole	Propiconazole 25%	0.5 ml/litre

3.16 Preparation of fungicidal solution:

Fungicide solution were prepared dissolving require amount of fungicides with plain water to get proper concentration in 2000 ml beaker or bottle. Bottles were labeled appropriately. Before using the prepared solution were shaken thoroughly.

3.17 Plant extract used

Five plants extract *viz*. Allamanda leaf extract, Neem leaf extract, Garlic extract, Ginger extract and Onion extract were used in the experiment (Fig. 1) Details of the plant extract used are given below:

Table 4. Particulars of plant extracts used in the experiment

Treatment	Common Name	Scientific name	Plant parts used	Dose used (ration)
T_7	Allamenda	*Allamanda cathartica*	Leaf	1:4
T_8	Neem	*Azadirachta indica*	Leaf	1:4
T_9	Garlic	*Allium sativum*	Clove	1:4
T_{10}	Ginger	*Zingiber officinale*	Rhizome	1:4
T_{11}	Onion	*Allium cepa*	Bulb	1:4
T_{12}	Plain water	-	-	-

A. Allamanda Plant

B. Neem Twig

C: Garlic Clove

D: Ginger Rhizome

E: Onion bulb

Fig 1. Different types of plant species used in the experiment

(A. Allamanda plant, B. Neem leaf, C. Garlic Clove, D. Ginger rhizome and E. Onion bulb)

3.18 Collection and preparation of plant extracts solution

Five plants extract namely Allamanda leaf, Neem leaf, Garlic, Ginger and Onion were used in this study. For extraction of juice, required amount of respective parts of each pant parts were taken, washed in tap water and crushed in a mortar and pestle. The crushed materials were blended in an electric blender adding double amount of water for 1: 2 solution. The blended materials were filtered through sterile cheesecloth. The supernatant was diluted in equal amount of sterile water for 1: 4 solutions. The extracts were labelled and shaken thoroughly before use.

3.19 Tagging

Five plants were selected randomly from each plot for determining of leaf infection, leaf area diseased, twig infection, fruit infection and fruit area diseased.

3.20 Application of fungicides and plant extracts

All fungicides and plant extracts were sprayed with compressed hand sprayer. Three plots were sprayed with each fungicides and plant extracts. First spray was done in 22 February, 2007. The plants under respective treatments were sprayed five times at 15 days intervals. Required amount of spray solution was applied per plant covering branches, leaves and fruits properly. Plants of control plot were sprayed with plain water only. Precautions were taken to avoid drifting of spray materials to neighboring plants with polythene barrier.

3.21 Diseased symptoms observed in the experimental plot

The plants were routinely observed from the time of spraying. Onset of new infection was recorded and symptoms of the anthracnose were observed, recorded and photographed.

3.22 Isolation of Pathogen

Infected leaves, twigs and fruits were collected for Isolation of the fungi. Stem piece, 1cm in length was cut out from the twigs for isolation. Four pieces of infected tissue approximately 10 mm in length and 1.5-2.5 mm width were surface sterilized in Chlorox (10%) solution for 45 seconds and washed thrice in sterile water. The inocula were then placed on acidified Potato Dextrose Agar (PDA) (Appendix.3) medium in pertri dishes aseptically. After planting, the petri dishes containing the inocula were incubated at room temperature ($26^0C \pm 2^0C$) under 12 hours light alternating with 12 hours dark. The plates were incubated for 7 days in the inoculation chamber

3.23 Identification of fungal isolates

Fungi growing out in the culture media were transferred to fresh PDA plates. The fungal isolates were then sub cultured on 2% water agar and purified by hyphal tip culture method. The fungus was identified following the appropriate keys (Kulsherestha *et al.*, 1979 and Sulton, 1980).

3.24 Data collection

Data were collected in the morning on the following parameters:

3.24.1.1 Total number of twigs/plant

Number of total twigs/plant was counted from randomly selected five plant from each plot at different dates as scheduled.

3.24.1.2 Total number of infected twigs /plant

Number of infected twigs/plant under each treatment was counted at different observation dates as scheduled.

3.24.1.3 Calculation of diseased incidence of twig anthracnose of different treatments

The percent diseased incidence of twig anthracnose was calculated using the following formula.

$$\text{Disease incidence} = \frac{\text{Number of infected twig}}{\text{Total number of observation (twig)}} \times 100$$

3.24.2.1. Total number of leaves/plant

Number of total leaves was recorded at different observation dates as scheduled.

3.24.2.2 Total number of infected leaves /plant

Number of anthracnose infected leaves/plant under each treatment was counted at different observation dates as scheduled

3.24.2.3 Calculation of diseased incidence of leaves caused by anthracnose of different treatments

The percent diseased incidence of was calculated using the following formula.

$$\text{Disease incidence} = \frac{\text{Number of infected leaves}}{\text{Total number of observation (leaves)}} \times 100$$

3.24.3.1 Total number of fruit/plant

Number of total fruit was recorded at different observation dates as scheduled.

3.24.3.2 Total number of anthracnose infected fruits/plant

Number of anthracnose infected fruits/plant under each treatment was counted at different observation dates as scheduled

Number of total fruit was recorded at different observation dates as scheduled.

3.24.3.3 Calculation of diseased incidence of fruits caused by anthracnose of different treatments

The percent diseased incidence of was calculated using the following formula.

$$\text{Disease incidence} = \frac{\text{Number of infected fruits}}{\text{Total number of observation (fruits)}} \times 100$$

3.25 Evaluation of leaf and fruit anthracnose severity

Percent leaf area diseased (LAD) and fruit area diseased (FAD) were measured. Area of a single leaf /fruit was considered as 100%. Deducting the healthy area, the diseased area was estimated. Average of % LAD and FAD was then calculated dividing the total disease areas by total number of investigated leaves/fruits (Islam *et al.*, 2001). The leaf and fruit anthracnose severity was recorded following 0-5 scale with slight modification as designed by Basak (1997) (Fig. 2).

The grade is given below

Grade		**% area infected**
0	=	No infection
1	=	Up to 5% area infected
2	=	5.1-12.0% areas infected
3	=	12.1-25.0% areas infected
4	=	25.1-50.0% area infected and
5	=	Above 50.1% area infected.

Fig 1. Diseased severity scale (0-5)

0 = 0% Fruit Area Diseased (FAD)

1 = 1-5% Fruit Area Diseased (FAD)

2 = 5.1 -12.0 Fruit Area Diseased (FAD)

3 = 12.1 -25. Fruit Area Diseased (FAD)

4 = 25.1 -50.0 Fruit Area Diseased (FAD)

5 = >50.1 Fruit Area Diseased (FAD)

3.26 Assessment of anthracnose of chilli

Five-plants/ plot were randomly tagged for recording infection. On the expression of the symptoms as leaf infection, fruit infection, lesion area on fruit was taken at 15 days interval after application of treatments. Total number of leaves or fruits and number of infected leaves or fruits was counted to calculate the percent leaf or fruit infection. The disease incidence was recorded using the methodology followed by Basak *et al.* (1990, 1991)

$$\% \text{ Leaf or fruit area infection} = \frac{\text{Number of infected leaves or fruits}}{\text{Total number of leaves or fruits}} \times 100$$

3.27 Isolation, purification and identification of the organism

Diseased fruit with typical symptom was collected for isolation of the organism in the laboratory. Potato Dextrose Agar (PDA) (Appendix 3.) media was prepared and transferred to glass petridishes. The fruit samples were cut into small pieces. Each pieces contained diseased and healthy tissue. Surface sterilization of the pieces with chlorox (1:1000) for 1 minute was done followed by three washes with sterile water and was placed onto PDA media in petri dish. The plates were incubated at 28±1°C for growth of the fungal colony.

Pure culture of the organism was prepared by means of hyphal tip culture method aseptically and repetition of sub-culture into PDA to at least 3-5 times. Colony characters, mycelial growth, color and sporulation of the fungi were studied and identified the organism following the methods of Singh (1982).

3.28 Yield of Chilli fruit

The weight of fresh and ripe fruits per treatment was recorded at the time of every harvest, where harvesting was done with four times. The weight of fruits per plot was recorded and converts into per hectare yield.

3.29 Cost Benefit Analysis and Calculation of benefit cost Ratio (BCR)

Costing of application of different fungicides and plant extracts for management of anthracnose of chilli was done based on the current market price of input, rate of hiring labor and agricultural machineries. Price of the field product was determined on the basis of current market value (Appendices 5 and 6). Estimation of Benefit Cost Ratio (BCR) was done according to Gittinger (1982) and Islam (2004) using the following formula-

$$BCR = \frac{\text{Gross return (Tk/ha)}}{\text{Total cost of production (Tk/ha)}}$$

3.30 Analysis of data

The data on various parameters were analyzed using analysis of variance to find out the variation obtained from different treatments. Compilation of the experimental data and analysis were done by the computer MSTAT-C program following the statistical procedures of Gomez and Gomez (1983). Treatment means were compared by DMRT (Duncan's Multiple Range Tests).

CHAPTER FOUR

RESULTS

CHAPTER IV

RESULTS

4.1 Symptoms of anthracnose of chilli as observed in the field

The symptoms of this disease appeared on ripened fruits and therefore sometimes the disease is called "Ripe fruit rot". Circular and sunken lesions with black margins appeared on the ripe-fruits. Pinkish masses of spores covered the sunken spot. In the advanced stage of the disease, the concentric markings with dark acervuli appeared on the affected parts. The spotted fruits dropped down prematurely and resulted heavy losses. The fungus also attacked the fruit (Fig. 3) and stem causing die back (Fig. 6) symptoms.

4.2 Isolation and Identification of the pathogen of Die back/ anthracnose diseases of chili

a. Isolation and identification of *Colletotrichum capsici* from diseased specimen.
b. Bioassay of plant extracts and fungicides against *Colletotrichum capsici*

Cup / Groove method: From a PDA plate three 5 mm discs of the medium were scooped from three places maintaining an equal distance from the centre by a sterilized disc cutter. One milliliter of plant extract / fungicides solution was put into each hole and the plates were stored overnight in refrigerator for diffusion of the input in the medium around the hole before resumption of fungal growth. The next day, one 5 mm

block of 5 days old fungal culture cut by sterilized disc cutter and was placed at the centre of the plate. The growth of acervulus of *Colletotrichum capsici* was recorded at 24 hr. interval until the control plates were filled in (Nene and Thaplial, 1993; McKeen *et al.*, 1986).

The culture grown in PDA medium from diseased fruit showed that mycelium developed as white colony but the growth was very slow. After 6 days, numerous dots like black acervuli began to form through out the colony showing concentric ring. The appearance of the spore mass was pinkish and they were abundant.

The pathogen, *Colletotrichum capsici* was identified based on the asexual reproductive structures as found under compound microscope. Acervuli were disc shaped, waxy, sub epidermal, typically with dark needle like septate setae; conidiophore short, simple; conidia hyaline, one celled, ovoid or oblong (Fig. 4).

4.3 Effect of different fungicides and plant extract on germination percentage of chilli seeds in laboratory condition

The effect of different fungicides and plant extracts were presented in table 4. Among the fungicides, the highest seed germination (72%) was recorded in case of Ridomil Gold which was closely followed by Bavistin 50 WP (68%) and Proud 250 EC (68%). The lowest seed germination was recorded in case of Tilt 250 EC (32%). Among plant extracts, Ginger and Neem both yielded jointly the highest seed germination (80%) followed by Garlic extract (72%) and onion extract (64%). Forty four percent (44%) seed germination was recorded in control.

4.4 Effect of different fungicides and plant extracts on germination percentage of chilli seeds in seedbed

Effects of different fungicides and plant extracts on germination percentage of chilli seeds were presented in the Table 5. Among the fungicides the highest germination percentage of seeds (69.16%) observed in case of Ridomil Gold (T_5) and the lowest germination percentage (28.33%) was observed in Tilt 250 EC (T_6). The second highest performance in seed germination was showed by Bavistin 50 WP (63.5%) which was closely followed by Cupravit 50 WP and Proud 250 EC. Among the plant extracts the highest seed germination percentage (78.33%) was observed in case of ginger extract (T_8) followed by Neem extract (74.0%) and garlic extract (68.66%). The lowest seed germination percentage (56.66%) was recorded in onion extract (T_{11}) which was also better than control (34.50%).

Table 4. Effect of different treatment on seed germination in lab condition

Treatment	No. of total seeds for plating	No. of germinated seed	Germination percentage (%)
T_1 (Bavistin 50 WP)	25	17	68
T_2 (Cupravit 50 WP)	25	16	64
T_3 (Dithane M 45)	25	12	48
T_4 (Proud 250 EC)	25	17	68
T_5 (Ridomil Gold)	25	18	72
T_6 (Tilt 250 EC)	25	08	32
T_7 (Allamanda leaf extract)	25	15	60
T_8 (Neem leaf extract)	25	20	**80**
T_9 (Garlic extract)	25	18	72
T_{10} (Ginger extract	25	20	**80**
T_{11} (Onion extract)	25	16	64
T_{12} (Control)	25	11	44

Table 5. Effect of different treatment on seed germination in field condition

Treatment	No. of total seeds sown	No. of germinated seed	Germination Percentage (%)
T_1 (Bavistin 50 WP)	200	127.0	63.5
T_2 (Cupravit50 WP)	200	125.6	62.8
T_3 (Dithane M 45)	200	87.66	43.83
T_4 (Proud 250 EC)	200	124.32	62.16
T_5 (Ridomil Gold)	200	138.32	69.16
T_6 (Tilt 250 EC)	200	56.66	28.33
T_7 (Allamanda leaf extract)	200	114.66	57.33
T_8 (Neem leaf extract)	200	148.0	74.00
T_9 (Garlic extract)	200	137.32	68.66
T_{10} (Ginger extract	200	157.66	**78.83**
T_{11} (Onion extract)	200	113.32	56.66
T_{12} (Control)	200	69.0	34.50

4.5 Effect of different fungicides and plant extracts on disease incidence and severity of anthracnose of chilli

Effect of different fungicides on percent leaf infection and per cent leaf area diseased (LAD) are presented in Table 6. The lowest leaf infection (4.52%) was recorded in case of spraying Proud 250 EC which was statistically similar to that of Cupravit 50 WP (4.55%) followed by Tilt 250 EC (4.74%) and Garlic extract (4.89%). The highest leaf infection (6.68%) was noted in control preceded by Ridomil Gold (5.54%) and Allamanda extracts (5.38%). Based on the performances of fungicides and plant extracts in controlling incidence of anthracnose of chilli, it was observed that highest reduction of leaf infection (32.32%) was shown by spraying with Proud 250 EC followed by Cupravit 50 WP (31.90%) and Tilt 250 EC (29.09%). Among the plant extracts Garlic showed the best performance in reduction of leaf infection (26.77%).

The performances of the fungicides and plant extracts in respect of percent Leaf Area Diseased (LAD) were differed significantly. The lowest score of Leaf Area Diseased (3.56%) was recorded in case of Proud 250 EC which was statistically similar with that of Allamanda leaf extract (3.8%). The second lowest LAD (4.1%) was noted in case of Tilt 250 EC which was also statistically similar with Bavistin 50 WP (4.33%). The highest percent of LAD (7.15%) was recorded in control. On the basis of percent LAD recorded in different treatments it was calculated that the highest reduction of LAD (50.20%) was found incase of Proud 250 EC followed by Allamanda leaf extract (46.85%), Tilt 250 EC (41.81%) and Bavistin 50 WP (39.44%). The performance of the rest of the fungicides and plant extracts were not found promising in reduction of disease severity.

ble 6. Effect of different fungicides and plant extracts on disease incidence and severity of anthracnose of chilli

Treatments	Leaf infection (%)	Reduction of leaf infection (%)	Leaf Area Diseased (% LAD)	Reduction of LAD (%)
T_1 (Bavistin 50 WP)	5.17 d	22.68	4.33 f	39.44
T_2 (Cupravit50 WP)	4.55 h	31.90	5.01 e	29.93
T_3 (Dithane M 45)	5.47 bc	18.12	5.16 e	27.83
T_4 (Proud 250 EC)	4.52 h	32.32	3.56 g	50.20
T_5 (Ridomil Gold)	5.54 b	17.10	5.25 de	26.57
T_6 (Tilt 250 EC)	4.74 g	29.09	4.16 f	41.81
T_7 (Allamanda leaf extract)	4.89f	26.77	3.80 g	46.85
T_8 (Neem leaf extract)	5.26 d	21.25	6.01 b	15.94
T_9 (Garlic extract)	5.04 e	24.51	5.08 e	28.95
T_{10} (Ginger extract	5.38 c	19.49	5.56 cd	22.23
T_{11} (Onion extract)	5.19 d	22.32	5.65c	20.98
T_{12} (Control)	6.68 a	----	7.15 a	--
CV (%)	4.41	----	14.40	----
DMRT(lsd)	0.105		.0312	

Means in a column having a common letter (s) do not differ significantly (P = 0.05) by DMRT. Data were analyzed after square root transformation.

4.6 Effect of different fungicides and plant extracts on die back (twig infection) of chilli

Effect of different fungicides and plant extracts on die back (twig infection) is presented in Table 7. The lowest twig infection (4.71%) of chilli was recorded in case of Proud 250 EC that was similar with that of Tilt 250 EC (4.78%). The highest twig infection (6.86%) was recorded in control treatment. The reduction of twig infection was 31.32% and 30.32%, respectively in case of spraying Proud 250 EC and Tilt 250 EC. The performance of the rest of the fungicides and plant extracts were not found promising.

ble 7. Effect of different fungicides and plant extracts on die back (twig infection) of chilli

Treatments	Die-back (% twig affected)	Reduction of die back (%)
T_1 (Bavistin 50 WP)	5.77 b	15.88
T_2 (Cupravit 50 WP)	5.43 cd	20.84
T_3 (Dithane M 45)	5.40 cd	21.28
T_4 (Proud 250 EC)	**4.71 f**	**31.34**
T_5 (Ridomil Gold)	5.06 e	26.24
T_6 (Tilt 250 EC)	4.78 f	30.32
T_7 (Allamanda leaf extract)	5.12 e	25.36
T_8 (Neem leaf extract)	5.56 bc	18.95
T_9 (Garlic extract)	5. 21 de	24.05
T_{10} (Ginger extract	5.27 de	23.17
T_{11} (Onion extract)	5.70 b	16.90
T_{12} (Control)	6.86 a	--
CV (%)	9.59	--
DMRT(lsd)	0.225	

Means in a column having a common letter (s) do not differ significantly (P = 0.05) by DMRT. Data were analyzed after square root transformation.

4.7 Effect of different fungicides and plant extracts on fruit infection and lesion Fruit Area Diseased (FAD)

Effect of different fungicides and plant extracts on fruit infection and fruit area diseased were summarized in Table 8. The treatments showed statistically significant effect on fruit infection and fruit area diseased (FAD). The highest percent of fruit infection was observed in control plot T_{12} (6.41%) and the lowest percentage of fruit infection was observed in Cupravit 50 WP (2.73%) which was statistically similar to that of Tilt 250 EC (2.86%) followed by Allamanda leaf extract(2.91%) and Proud 250 EC (3.16%). Based on the performance against fruit infection the highest reduction of fruit infection (57.41%) was found incase of Cupravit 50 WP followed by Tilt 250 EC (55.35%), Allamanda extracts (54.62%) and Proud 250 EC (50.64%) in comparison to control.

Incase of Fruit Area Diseased (FAD), the lowest severity (2.97%) and highest reduction of severity (42.44%) was recorded in case of Cupravit 50 WP and Tilt 250 EC. The second highest severity (3.51% FAD) was noted in Proud 250 EC that was statistically similar to Ridomil Gold (3.52% FAD). The Allamanda leaf extract also showed better performance in reduction of % FAD in comparison to control.

Table 8. Effect of different fungicides and plant extracts on fruit infection and fruit area diseased (FAD)

Treatments	Fruit infection (%)	Reduction of fruit infection (%)	Fruit Area Diseased (% FAD)	Reduction of Fruit area diseased (%)
T_1 (Bavistin 50 WP)	3.27 ef	48.97	4.40 c	14.72
T_2 (Cupravit 50 WP)	2.73 h	57.41	2.97 g	42.44
T_3 (Dithane M 45)	3.97 b	38.08	3.96 de	23.25
T_4 (Proud 250 EC)	3.16 f	50.64	3.51 f	31.97
T_5 (Ridomil Gold)	3.36 de	47.59	3.52 f	31.78
T_6 (Tilt 250 EC)	2.86 gh	55.35	2.97 g	42.44
T_7 (Allamanda leaf extract)	2.91 g	54.62	3.80 e	26.36
T_8 (Neem leaf extract)	3.44 d	46.35	4.00 de	22.48
T_9 (Garlic extract)	4.07b	36.51	3.96 de	23.25
T_{10} (Ginger extract	3.95b	38.57	4.65 b	9.88
T_{11} (Onion extract)	3.58 c	44.05	4.91 cd	4.84
T_{12} (Control)	6.41 a	---	5.16 a	----
CV (%)	8.46		13.04	
DMRT(lsd)	0.137		0.226	

Means in a column having a common letter (s) do not differ significantly (P = 0.05) by DMRT. Data were analyz after square root transformation

4.8 Effect of different fungicides and plant extracts on fruit yield against anthracnose of chilli

Effect of different fungicides and plant extracts on yield of chilli against anthracnose disease was determined and presented in Table 9. The effect of different fungicides and plant extracts on fruit yield of chilli against anthracnose was differed significantly (Table. 10). The highest yield (10.83 t/ha) was recorded in the plot treated with Tilt 250 EC (Fig. 7) which was statistically identical with that of Proud 250 EC (10.16 t/ha). Bavistin 50 WP (9.5 t/ha), Dithane M- 45 (8.66 t/ha) and Cupravit 50 WP (8.53 t/ha) also showed better performance on fruit yield against anthracnose of chilli. The performance of plant extracts in contributing fruit yield of chilli against anthracnose disease was not up to the mark except Allanamda (8.41 t/ha). On the basis of yield performance of the treatments applied against anthracnose of chilli, it was observed that the highest increase (195.90 %) of fruit yield was achieved by spraying Tilt 250 EC followed by Proud 250 EC (177.59 %) and Bavistin 50 WP (159.56 %) compared to control. Cupravit 50 WP, Dithane M-45 and Allamanda extract also showed better performance in increasing fruit yield of chilli against anthracnose disease.

Table 9. Effect of different fungicides and plant extracts on fruit yield of chilli against anthracnose disease

Treatments	Yield (ton/ha)	% increase of production
T_1 (Bavistin 50WP)	9.50 bc	159.56
T_2 (Cupravit 50WP)	8.53 d	133.06
T_3 (Dithane M-45)	8.66 cd	136.61
T_4 (Proud 250EC)	10.16 ab	177.59
T_5 (Ridomil Gold)	6.35 e	73.49
T_6 (Tilt 250EC)	10.83a	195.90
T_7 (Allamanda leaf extract)	8.41 d	129.78
T_8 (Neem leaf extract)	7.18 e	96.17
T_9 (Garlic extract)	4.76 f	30.05
T_{10} (Ginger extract	4.2 g	14.75
T_{11} (Onion extract)	5.24 f	43.17
T_{12} (Control)	3.00g	--
CV (%)	25.92	--
DMRT (lsd)	0.891	

Means in a column having a common letter (s) do not differ significantly (P = 0.05) by DMRT. Data were analyzed after square root transformation

4.9 Cost analysis

The result of cost analysis has been presented in the Table 10. Estimation showed that some treatments had tremendous performance regarding BCR (Benefit Cost Ratio). The highest BCR (4.07) was observed in T_6 (Tilt 250 EC) followed by Proud 250 EC (3.68), Bavistin 50 WP (3.47) and Allamanda extract (3.16). The Benefit Cost Ratio (BCR) of the application of garlic extract (1.34), ginger extract (1.23) and onion extract (1.59) were not economically applicable in comparison to control (1.20).

Table 10. Cost Benefit analysis of twelve different treatments for controlling anthracnose of chilli

Treatment	Average yield (Ton/ha)	Gross return Tk./ha	Total cost (Tk./ha)	Gross margin	BCR
T_1 (Bavistin 50 WP)	9.50	190000	54720	135280	3.47
T_2 (Cupravit 50 WP)	8.53	170600	61920	108680	2.75
T_3 (Dithane M 45)	8.66	173200	58652	114548	2.95
T_4 (Proud 250 EC)	10.16	203200	55120	148080	3.68
T_5 (Ridomil Gold)	6.35	127000	56420	70580	2.25
T_6 (Tilt 250 EC)	10.83	216600	53170	163430	4.07
T_7 (Allamanda extract)	8.41	168200	53220	114980	3.16
T_8 (Neem extract)	7.18	143600	53220	90380	2.69
T_9 (Garlic extract)	4.76	95200	70720	24480	1.34
T_{10} (Ginger extract	4.20	84000	68220	15780	1.23
T_{11} (Onion extract)	5.24	104800	65720	39080	1.59
T_{12} (Control)	3.00	60000	49620	10380	1.20

Fig 3. Severely affected fruits caused by *Colletotrichum capsici*.

Fig.4. Acervulus with setae, conidiophores and conidia of *Colletotrichum capsici* seen under compound microscope (10X)

Fig 5. A view of the field experiment of chilli at the farmland of Sher-e-Bangla Agricultural University

Fig 6: A view of the affected plot (control) showing die back symptom of chilli

Fig 7. Healthy plot of chilli treated with Tilt 250 EC against anthracnose of chilli

CHAPTER FIVE

DISCUSSION

CHAPTER V

DISCUSSION

Anthracnose caused by *Colletrotichum capsici* is a serious disease of chilli (Pillayrsany *et al.*, 1973). The disease affects fruits resulting in lower yield and poor quality fruits (Chowdhury, 1975). Symptoms of the disease as observed in the present study confirm to those described by Basak *et al.* (1991)

Different fungicides and plant extracts showed significant variation in performance against *Colletrotichum capsici* causing anthracnose of chilli. On the basis of the overall effectiveness of the fungicides against the disease, Tilt 250 EC and Proud 250 EC found to be the effective fungicides. Proud 250 EC reduced 50.20% leaf area diseased (LAD) while Tilt 250 EC reduced 41.81% LAD. Among rest of the fungicides Bavistin 50 WP showed better performance in reduction of LAD (39.44%) against the disease. In case of twig infection, the highest reduction of die back also scored by Proud 250 EC (31.34%) followed by Tilt 250 EC (30.34%). Almost similar performance of the fungicides was noticed in fruit infection and reduction of fruit area disease (FAD). The lowest infection of fruit were observed in Tilt 250 EC (2.86%) and Cupravit (2.73%) followed by Proud 250 EC (3.16%). Simultaneously, the highest reduction of FAD (42.44%) was noted in Tilt 250 EC and Cupravit 50WP that was followed by Proud 250 EC (31.97%). The findings of the present investigation corroborated with the findings of Ahmed (1986), Eawaramurthy (1988), Rahman *et al.* (1994), Ebenezar and Alice (1996), Hedge and Anahosur (2001), Deeksha (2002), Anon. (2005) and Roy (2005). In a similar type of study, Eawaramurthy (1988) found that copper oxychloride (Cupravit 50

WP) @ 0.25% proved to be potential in controlling die back of *Capsicum annuum* increasing fruit yield. Rahman *et al.* (1994) reported that Tilt 250 EC had promising effect in *in vivo* and *in vitro* against *Colletrotichum lindemuthianum* and *Colletrotichum dematium*. Hedge and Anahosur (2001) while evaluating non systemic and systemic fungicides against anthracnose of chilli reported the fungicides of group Propicanozole and carbendazim group should effective performance in controlling fruit rot of chilli. Roy (2005) reported while working on the efficacy of fungicides against anthracnose of chilli that Tilt 250 EC (Propicanozole) proved to be effective against die back and anthracnose of chilli. According to Anon. (2005) it was reported that Tilt 250 EC was found effective in controlling anthracnose of chilli. Deeksha *et al.* (2002) also reported that Bavistin 50 WP and Tilt 250 EC (Propicanozole) @ 0.1% gave good control of anthracnose of *Vigna mungo*.

Considering the overall performance of the plant extracts against anthracnose of chilli, Allamanda leaf extract reduced leaf infection, twig infection and fruit infection by 26.77%, 25.36% and 54.62%, respectively. Reports on allamanda extract against anthracnose of chilli are not available in the literature. However, evidences of using allamanda leaf extracts against other fungal pathogens were available in literature (Howlader, 2003 and Islam, 2004). Howlader (2003), working with plant extracts and fungicides against *Phomopsis vexans*, found that allamanda leaf extract had profound effect against *Phomopsis vexans* causing Phomopsis blight of egg plant. Islam, (2004) also reported that allamanda leaf extracts caused 76-100% inhibition of mycelial growth of *Phomopsis vexans*. She further reported that TLC studies of the allamanda extract showed that the presence of a number of compounds having very low to high polarity might contribute to inhibit the pathogen. Garlic extracts reduced leaf, twig

and fruit infection by 26.77%, 24.05% and 36.51%, respectively. Meah (2003) and Arun *et al.*, (1993) reported that garlic extract was the most effective against anthracnose of chilli. Singh *et al.*, (1997) reported that garlic has antifungal activity against the anthracnose pathogen of chilli (*Colletotrichum capsici)* and caused 100% inhibition of mycelial growth and spore germination. The present findings are also well supported by the findings of Harbant *et al.*, (1999) who reported that garlic extracts controlled anthracnose of chilli significantly over control.

Cost analysis showed that application of Tilt 250 EC scored the highest BCR (4.07) where Tk. 3.07 could be earned investing Tk. 1.00. The second highest BCR (3.68) was achieved by Proud 250 EC followed by Bavistin 50 WP (3.47) and allamanda extracts (3.16). The lower BCR in case of garlic extract indicated that the investing cost of garlic is comparatively higher than other extracts.

From the findings of the present investigation, it may be concluded that Propiconazole fungicides (like Tilt 250 EC or Proud 250 EC) or carbendazim like Bavistin 50 WP could be used in controlling anthracnose of chilli. Among in plant extracts use of allamanda extract would be economically viable than other plant extract against anthracnose of chilli.

CHAPTER SIX

SUMMARY AND CONCLUSION

CHAPTER VI

SUMMARY AND CONCLUSION

An experiment was conducted at the farm of Sher-e-Bangla Agricultural University during the period of October'06-May'07 to find out the effective fungicides and plant extracts in controlling anthracnose of chilli. Six fungicides *viz.* Bavistin 50 WP, Cupravit 50 WP, Dithane M- 45, Proud 250 EC, Ridomil Gold and Tilt 250 EC and five plants extracts *viz.* Allamanda, Neem, Garlic, Ginger and Onion extracts were applied in the experiment.

The effect of fungicides and plant extracts in controlling the anthracnose of chilli were determined by recording data after different days of spraying in terms of disease incidence disease severity and yield. The effects of treatments were compared by Duncan's Multiple Range Test (DMRT).

After 75 days, after starting of spray (five sprays has been given at 15 days intervals) the lowest incidence of fruit infection (2.73%) was found in Proud 250 EC (T_4) and the highest incidence of fruit infection of was found in control (T_{12}). The lowest incidence of LAD was found in Proud 250 EC (3.56%). The second lowest incidence of %LAD was found with Tilt 250 EC (4.16%) and Cupravit 50 WP (5.01%). The highest leaf infection (6.69%) and %LAD (7.15%) were recorded in case of control treatment.

After 75 days, after starting of spray the lowest incidence of twig infection (4.71%) was found in Proud 250 EC (T_4) that was statistically indifferent with that of Tilt 250 EC (4.78%). The highest fruit infection (6.68%) was

recorded in control treatment. The twig infection was reduced by 31.32% and 30.32% respectively for Proud 250 EC and Tilt 250 EC.

In case of fruit infection statistically the lowest incidence was recorded in Cupravit 50 WP (2.73%) and Tilt 250 EC (2.86%) that reduced 57.41% and 55.35% fruit infection, respectively over control. Similarly, the lowest %FAD was found in Cupravit 50 WP (2.97%) and Tilt 250 EC (2.97) that reduced 42.44% FAD in comparison to control. Proud 250 EC also showed remarkable performance in reducing %FAD (31.97%) over control.

After 75 days, after starting of spray, the highest yield of fresh fruit (10.83 ton/ha.) was found in Tilt 250 EC (T_6) and the second highest yields of fresh fruit were found in Proud 250 EC (T_4). The lowest yield of fresh fruit (3.00 ton/ha.) was found in control plot (T_{12}).

Regarding Benefit Cost Ratio (BCR), the highest BCR (4.07) was noticed in case of Tilt 250 EC followed by Proud 250 EC (3.68), Bavistin 50 WP (3.47) and Allamanda extract (3.16). Cost analysis showed that use of Garlic, Ginger and Onion extract did not found economically viable.

Considering the overall performance of the treatments applied, it is revealed that Tilt 250 EC and Proud 250 EC showed promising performance in controlling anthracnose of chilli as well as increasing yield. However, further investigation need to be carried out in different Agro. Ecological Zones of Bangladesh for consecutive years for confirmation of the present findings.

CHAPTER SEVEN

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CHAPTER VII
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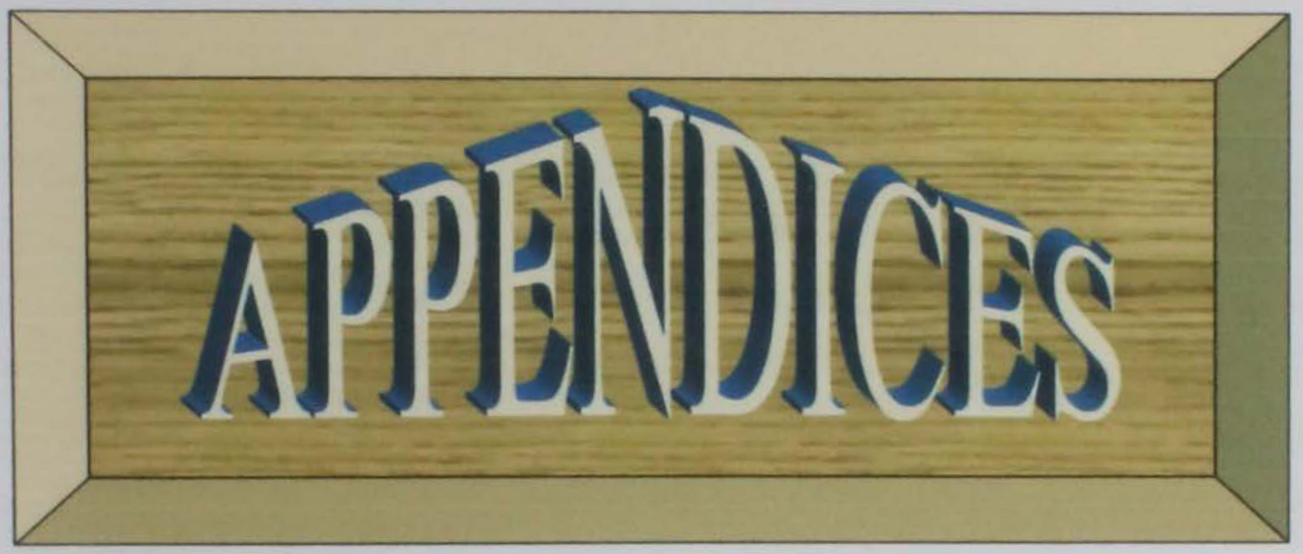

APPENDICES

APPENDICES

Appendix 1. Result of mechanical and chemical analysis of soil of the experiment plot

Mechanical analysis

Constituents	Per cent
Sand	33.45
Silt	60.25
Clay	6.20
Textural class	Silty lome

Chemical analysis

Soil properties	Amount
Soil	6.12
Organic matter	1.32
Total nitrogen (%)	0.08
Available P (ppm)	20
Exchangeable K (%)	0.2

Appendix 2. Monthly average temperature, relative humidity and total rainfall of the experimental site during the period from October 2006 to May 2007

Month	Air temperature (°C)		Relative Humidity (%)	Total Rainfall (mm)
	Maximum	Minimum		
October'06	26.70	21.13	89	41
November'06	22.00	20.15	87	00
December'06	20.00	20.90	64	00
January'07	24.90	13.20	67.5	3.0
February'07	28.10	17.80	61.5	4.0
March'07	32.50	22.60	66	155
April'07	33.50	23.80	85	140
May'07	34.30	24.40	77	165

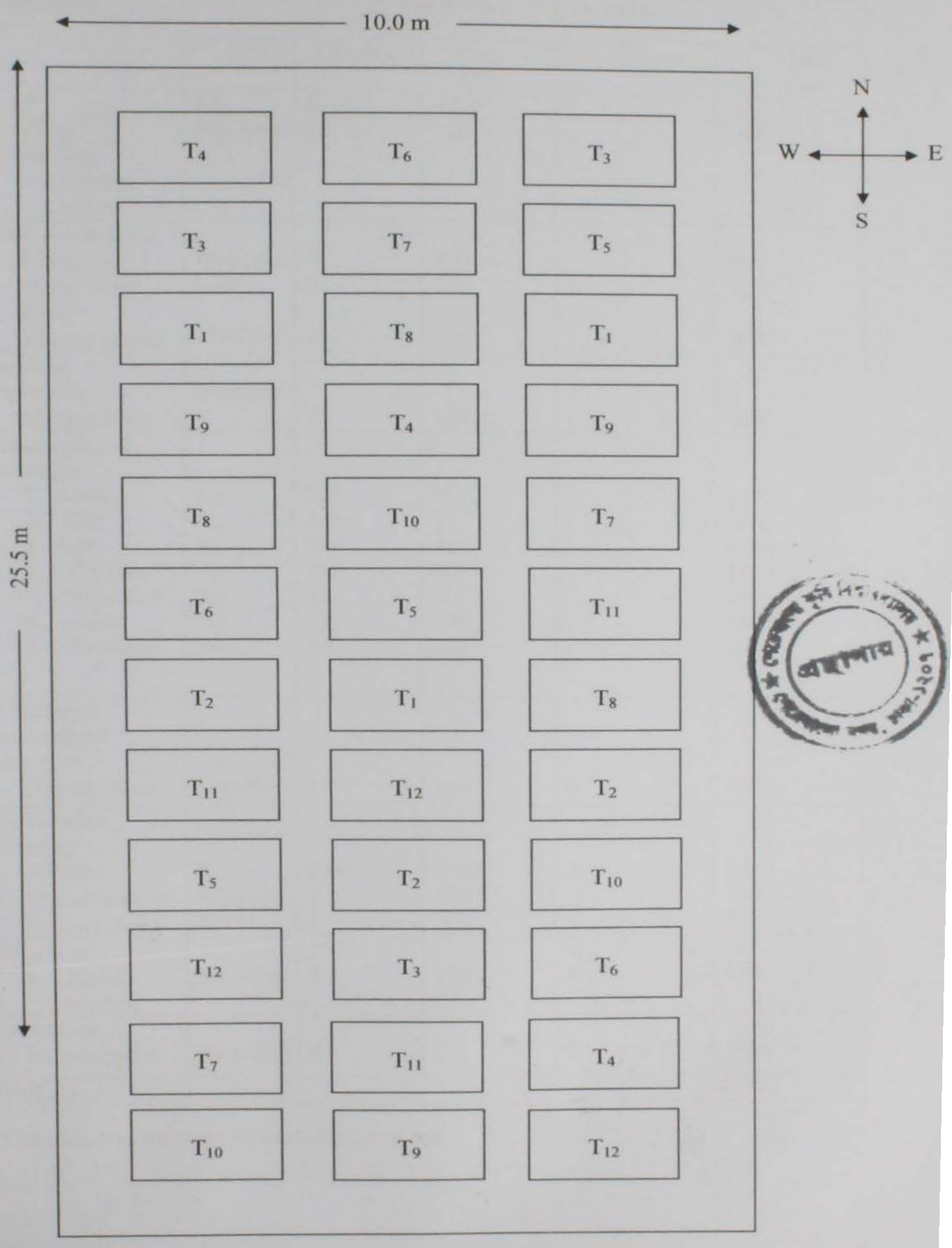
Source: Dhaka metrological centre

Appendix 3. Composition of potato Dextrose Agar (PDA)

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

Appendix 4. Design of the experimental plot

Plot size = 2.0 m × 1.5 m
Plot spacing = 50 cm
Between replication = 1.0 m



Appendix 5. Analysis of cost of application of common culture practice in production of chilli plant

Cost items	Per hectare cost in Taka				
	Unit	Quantity	Cost /unit	Times	Total Cost
Seed	Kg	0. 5	1000	-	500
Seedling production • Human labour	Man/day	12	120	-	840
Land preparation • Ploughing • Power tiller hired • Human labour	 Bull pair Hour Man/day	 5 6 25	 250 500 120	 - - -	 500 3000 3000
Seedling plantation • Human labor	Man/day	25	120	-	3000
Fertilization & Manuring 1. Urea 2. TSP 3. MP 4. Gypsum 5. Boric acid 6. $ZnSO_4$ 7. Cowdung	 Kg Kg Kg Kg Kg Kg Ton	 210 300 200 110 05 15 10	 07 24 20 10 120 120 500	 - - - - - - -	 1470 7200 4000 1100 600 1800 5000
Weeding & intercultural operation • Human labor	Man/day	15	120	4	7200
Insecticides Spraying • Aktara • Human labour • Sprayer hired	 Kg Man/day Hour	 0.60 2 3	 7000 120 50	 3 3 3	 4200 720 450
Irrigation • Human labour	Man/day	3	120	4	1440
Harvesting • Human labor	Man/day	6	120	5	3600
Total (a)					49620

***Calculated on the basis of market price of 2007.**

Appendix 6. Analysis of cost of application for management of Anthracnose of chilli

Cost items	Per hectare cost in Taka				
	Unit	Quantity	Cost/unit*	Times	Totalcost
Bavistin 50 WP	1 packet	10.0	66	5	3300
• Chemical	=50gm				
• Labor	Man/day	3	120	5	1800
Total (b)					5100
Cupravit 50 WP	1 packet	35	60	5	10500
• Chemical	=100gm				
• Labor	Man/day	3	120	5	1800
Total (c)					12300
Dithane M-45	1 packet	5	65	5	7232
• Chemical	=100gm				
• Labor	Man/day	3	120	5	1800
Total (d)					9032
Proud 250 WP	1 Bottle =	4	185	5	3700
• Chemical	125 ml				
• Labor	Man/Day	3	120	5	1800
Total (e)					5500
Ridomil Gold	1 Packet =	10	100	5	5000
• Chemical	100 gm				
• Labor	Man/day	3	120	5	1800
Total (f)					6800
Tilt 250 EC	1 Bottle =	2	175	5	1750
• Chemical	125 ml				
• Labor	Man/Day	3	120	5	1800
Total (g)					3550
Allamenda leaf extract					
• Labor	Man/Day	6	120	5	3600
Total (h)					3600
Neem leaf extract					
• Labor	Man/Day	6	120	5	3000
Total (i)					3600
Garlic extract					
• Garlic Clove	Kg	100	30	5	17500
• Labor	Man/Day	6	120	5	3600
Total (j)					21100
Ginger extract					
• Ginger rhizome	Kg	100	35	5	15000
• Labor	Man/Day	6	120	5	3600
Total (k)					18600
Onion extract					
• Onion bulb	Kg	100	25	5	12500
• Labor	Man/Day	6	120	5	3600
Total (l)					16100

***Calculated on the basis of market price of 2007**