

**MANAGEMENT OF *CHILLI LEAF CURL VIRUS* (ChLCV) OF CHILLI USING
SELECTED BOTANICALS, RAW COW MILK, BIOAGENT AND INSECTICIDE
UNDER FIELD CONDITION**

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CERTIFICATE

This is to certify that thesis entitled “**MANAGEMENT OF *CHILLI LEAF CURL VIRUS (ChLCV) OF CHILLI USING SELECTED BOTANICALS, RAW COW MILK, BIOAGENT AND INSECTICIDE UNDER FIELD CONDITION***” submitted to the Department of Plant Pathology, Sher-e- Bangla Agricultural University (SAU), 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 **TABASSUM TAMIMA**, Registration No. **14-06294** under my supervision and guidance. No part of the thesis has been submitted for any other degree or diploma.

I further certify that any help or source of information, received during the course of this investigation has been duly acknowledged and style of this thesis have been approved and recommended for submission.

Dated: June, 2021
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**Dedicated To
My Amicable Family**

MANAGEMENT OF *CHILLI LEAF CURL VIRUS* (ChLCV) OF CHILLI USING SELECTED BOTANICALS, RAW COW MILK BIOAGENT AND INSECTICIDE UNDER FIELD CONDITION

ABSTRACT

Chilli (*Capsicum frutescens*) is one of the most important spice and vegetable crop in the world which belongs to Solanaceae family. A field experiment was conducted at the central field of Sher-e-Bangla Agricultural University, Dhaka, Bangladesh during December 2020 to May 2021. The study was aim to assess the disease incidence (%) and the percent disease index of *Chilli leaf curl virus* under natural field condition by using different treatments through vector control activities. In this study, total 9 treatment were used viz. T₁ (Neem leaf extract), T₂ (Mahogoni leaf extract), T₃ (Garlic extract), T₄ (Trichoderma +Raw cow milk), T₅ (Malathion @ 3ml/L), T₆ (Lantana leaf extract), T₇ (Raw cow milk), T₈ (Trichoderma), T₉ (Marigold leaf extract) and one without treatment served as control (T₀). The field experiment was carried out in randomized complete block design (RCBD) with three replications. Among all the treatments, the minimum disease incidence, were recorded in T₅ (Malathion) and T₁ (Neem leaf extract) which were showed better performance aganist ChLCV. A positive correlation was found between white fly population and disease incidence as well as yield loss and white fly population. Infection by chilli leaf curl disease adversely affected growth and yield attributing characters. However, these two treatments T₅ (Malathion) and T₁ (Neem leaf extract) may be considered as better option to manage ChLCV through vector control.

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LIST OF ACRONYMS

Acronyms	Full form
AEZ	Agro-Ecological Zone
BARI	Bangladesh Agricultural Research Institute
BBS	Bangladesh Bureau of Statistics
<i>et.al</i>	And others
TSP	Tripple Supper Phosphate
MOP	Muriate of Potas
DAT	Days after transplanting
Kg	Kilogram
t/ha	Ton per hectre
%	Percentage
CV%	Percent of coefficient value
LSD	Least significant difference
Cm	Centimeter
SAU	Sher-e-Bangla Agricultural University
SRDI	Soil Research and Development Institute
FAO	Food and Agricultural Organization
M.S.	Master of Science
m ²	Meter squares

CHAPTER I

INTRODUCTION

Chilli is an important commercial spice as well as vegetable crop which is used worldwide for different purpose. Chilli (Marich) pod of a bushy plant, *Capsicum frutescens*, of the family Solanaceae, widely used as spice, native of Mexico. Capsicum is also known as chilli usually spelt 'chilli' in the United States, paprika, pimiento and sweet, red, cayenne or bird pepper depending upon the type and the way in which it is used vitamin. Capsanthin is the most important pigment of Capsicum. The pungent principle is Capsaicin, which is present in the placenta, and is said to retain its pungency in a dilution of one part in one million.

The spice crop chilli (*Capsicum annum L.*), known to be originated in tropical Asia, is cultivated in Bangladesh as the most important spice crop both during winter and summer seasons. In Bangladesh the crop is grown in an area of about 66,235 ha and annual production amounts to about 52,215 m.tons. According to present statistics, about 1, 50,000 ha land is under chilli cultivation in Bangladesh during the Rabi season and total production is about 2, 03,000 m.tons per year, approximately and per ha yield is about 1.35 m. tons. During the Kharif season about 35,000 ha of land is brought under chilli cultivation and total production is around 56,000 m.tons per year approximately and per ha yield is 1.59 m. tons. Chilli is a good source of vitamins and phosphorus. Chilli contains 1.29 mg protein, 11 mg calcium A, 17.5 mg ascorbic acid, 0.06 mg thiamine, 0.03 mg riboflavin and 0.55 mg niacin per 100 g edible green fruit (Joshi and Singh, 1975).

In Bangladesh many factors are responsible for the low yield of the crop. So far,65 viruses have been reported, including begomoviruses infecting chilli throughout the world (Nigam *et al.*, 2015).

Among all of these disease, viral infections are known to cause different symptoms for example mosaic, ring spot, curling, yellowing etc. on chilli and all these symptoms result in heavy losses. There are some agents like thrips, mites and whiteflies have been reported to produce viral disease and loses. Chilli are affected by 42 viruses. Among them, twenty of them reported to occur naturally and rest of them can infect on artificial inoculation.

24 viruses occur naturally on chilli, eleven viruses have been reported from India, viz., *Cucumber mosaic virus* (Anjaneyulu and Apps Rao,1967), *Tobacco leaf curl virus* (Hussam, 1932; and Vasudeva,1954), *Indian chilli mosaic virus* (McRae,1924 and Kha and Rayachaudhuri ,1956), *Potato virus Y* (Jeyarajan and Ramakrishna,1961 and 1969; and Rao *et al.*,1970). Prasad Rao (1976) reported that *Tabacco ring spot virus*, *Pepper venial mottle virus* and *Pepper vein banding virus*, *Chilli leaf curl virus* (Senanayake *et al.*, 2006) and *Tomato leaf curl New Delhi virus* (Ilyas, 1996). Among them, Chilli leaf curl disease (ChLCV) is occurred by (*Chilli leaf curl virus*) is a major limiting factor for chilli production in Indian subcontinent and begomo viruses is responsible for this disease (Bridoon *et al.*, 2003 ; Senanayake *et al.*, 2006; Shihab *et al.*, 2003) .

The chilli leaf curl disease on chilli was first reported in Pakistan by Shiv *et al.* (2003) and in India by Senanayake *et al.*,(2006). Leaf crinkle or leaf curl complex was observed on chillies by Hussain (1932). Leaf curl due to thrips results upward curling of leaves and interveina buckling. Irregular scraping of epidermis could also be seen (Johnpulie,1939).

The largest and most economically significant groups of plant virus transmitted by *Bemisia tabaci* are the Gemini viruses. It can be transmitted by mechanically, by vector, by seed. Several workers reported that the transmission of number of strains of TMV by mechanical sap inoculation chilli (Palm,Beecher, 1942;Lobachevsky, 1940, McKinney, 1952; Newton, 1954 ; Miller and Thornberr, 1958, Mura, 1923; Holmes, 1937; Namata and Takimoto, 1940; Doolittle and kshi, 1960 : Green leaf *et al.* ;1964; Ads uar etc al.,1971 Ragozzino *et.al.*,1972 ; Bid ari,1982). Doolittle and walker (1923), Doolittle and Zaumey'er (1953) reported CMV was mechanically sap transmitted. The leaf curl or leaf crinkle occurring on chillies was caused by *Bemisia tabaci* (*Bemisia gossypiperda*) was reported by Hussam. Mishra *et al.*, (1963), Muniyappa and Veeresh (1984) reported the transmission of chilli leaf curl by the victory called whitefly (*Besimia tabaci*). After 2-6 weeks, inoculated chilli plant showed typical leaf curl symptoms. Actually, the whitefly is the potential vector for the spread of *Chilli Leaf Curl Virus* which is causing serious damage to chilli crop. Whitefly sucks cell sap, secreting the honey dews and transmits a number of viral diseases (Khan and Ahmed, 2005). Hence, the infestation which is caused by whitefly

can cause severe and also transmitted *Chilli Yellow Leaf Curl virus* performed transmission by it and also crop damage can be reached up to 100% .

Senanayake *et al.* (2006) reported that a very high disease incidence (upto 100% plants during December, 2004) in farmers' fields in Narwa and Tinwari villages at Jodhpur district Rajasthan was observed. Chilli leaf curl disease complex causes huge crop losses in Jharkhand state primarily due to attack of thrips, mites and white fly followed by invasion of chilli leaf curl virus. ve viral pathogen in many parts of India, which is affecting chilli cultivation in terms of incidence and yield loss (Khan *et al.*, 2006).

Farmers regarded their economic losses from pests and diseases to be very substantial. Only a minority of them knew that certain disease symptoms were probably being caused by a plant virus and even fewer knew about the role of insect vectors in its spread. Farmers mostly relied on synthetic pesticides to manage the virus disease symptoms they observed. If farmers had better knowledge about plant viruses, their insect vectors, and cost-effective, safer means of control, then use of synthetic pesticides could be reduced substantially. Building knowledge among farmers is therefore an important way to address the diseases caused by plant viruses, while the development of virus-resistant varieties and simple and effective methods of vector control offer longer-term solutions.

The advent of synthetic insecticides in the mid-20th century made the control of insects and other arthropod pests much more effective, and such chemicals remain essential in modern agriculture despite their environmental drawbacks. By preventing crop losses, raising the quality of produce, and lowering the cost of farming, modern insecticides increased crop yields by as much as 50 percent in some regions of the world in the period 1945–65. They have also been important in improving the health of both humans and domestic animals; malaria, yellow fever, and typhus, among other infectious diseases, have been greatly reduced in many areas of the world through their use. (Britannica, Encyclopaedia. 2018).

Insecticides are applied to the environment with the aim of suppressing the impact of plant and animal pests and to protect agricultural and industrial products. For sustainable agriculture and protection of the environment and human health, the importance of using safe insecticides has assumed global importance subsequent to

the 'Earth Summit' in 1992. However, majority of the insecticides act like blunt weapons that kill organisms, in addition to target pests. Many of these substances are not easily degradable; they linger in soils, leach to ground water and contaminate the environment to a great extent. They can also enter the body of organisms, bioaccumulate in the food chains and consequently affect the human health. There exists a direct relationship between the extent of insecticides used and signs and symptoms of illness due to exposure among farmers (Kishi *et al.*, 1995).

On the other hand, if we use ecofriendly management to control any plant disease, our environment will be rescue from the adverse effect of chemical insecticides or pesticides.

For all these reasons, recent emphasis is on the development of non chemical / ecofriendly method for the management of leaf curl viral disease, which has provided impetus to more extensive exploration of natural resources and to identity effective plant extracts/botanical bio pesticides for the managements of chilli leaf curl virus by reducing vector population in the chilli plant. Indiscriminate use of insecticides created unwanted human health hazards. The current trends in plant pathology intend to boost the immunity of the host to increase the resistance against pathogens. The objective of this study are -

- To assess the disease incidence and percent disease index of Chilli leaf curl disease under different treatments and
- To evaluate the effect of different treatments on vector activities to reduce the disease intensity.

CHAPTER II

REVIEW OF LITERATURE

Chilli belongs to solaneaceae family which is a kind of bushy plant widely used as spice, native of Mexico. It is well known for its pleasant aromatic flavour pungency and high colouring substance having a medicinal value but there are 65 different diseases which affect chilli plant. The major constrain for the lower yield of chilli in our country is considered Chilli Leaf Curl Disease. Hence in this chapter an attempt was made to compile the relevant reviews for the present investigation.

2.1. Origin and Distribution

This is by far the most commonly collected Capsicum species in Africa. It is now found wild or as a weed from southern US through Mexico to N & E South America, the Antilles and Argentina. Delitt & Bosland (1996) suggested that it was first domesticated in Panama from where it spread to Mexico and the Caribbean. The domesticated variety of this species is usually known as var. pendulum, while the wild forms are known as var. baccatum and var. tomentosum and are commonly referred to as bird peppers. Many authors report the difficulty of differentiating herbarium specimens of this species from *C. annuum*. Bailey (in Man. Cult. Plants: 873 (1966)) considered both *C. annuum* and *C. baccatum* to be synonymous with *C. frutescens*, within which he differentiated several varieties often on their fruit size and shape.

It is also reported that chilli was originated in the American tropics and it has been propagated throughout the world which included the tropics, subtropics and also temperate regions (Pickers gill,1997).

In the year 2017, world production of fresh green chillies and peppers was 33.2 million tonnes and Bangladesh contributes 5% of the global total (FAOSTAT,2017)

In Bangladesh, it is cultivated in both the summer and winter seasons and average yield of green chilli is 1.3 ton/ha (BBS, 2018)

2.2. Nutritional value of chilli

Chilli is the member of solaneaceae which is reported to be rich in proteins, lipids, fibers, minerals salts, vitamins and in capsaicin (Chigoziri and Ekefan, 2013) For the sensory attributes pungency and aroma, chillies are most popular species in many parts of the world. In case of green chilli, it contains more vitamin C than citrus fruits and on the other hand, red chilli contains more vitamin A than carrots (Chigoziri and Ekefan 2013). Immature chilli fruit contain the phyto-nutrients ascorbic acid, carotenoids and rutin (Purseglove, 1975) Moreover, Chilli is suitable for not only diet of obese but also use to control cancer of stomach and colon (Dang *et al.*, 2014). Chilli fruits are used in sauces, soups, stews and generally as a flavouring agent and low in sodium and free cholesterol (Miller *et al.*, 2011) Capsicum in a fresh state is very rich in vitamin C, as cited by Dr. Szent Gyorgyi, the Hungarian scientist, who was awarded the nobel Prize in 1937 for isolating vitamin C from paprika fruits and showing that they were one of the richest sources available of this vitamin.

2.3. Diseases of Chilli

Chilli is suffering from about 65 different diseases (Nigham *et al.*,2015) .

Chilli is susceptible to various pathogens like viruses, fungi, bacteria which cause heavy production losses. So far, there are 65 viruses have been reported throughout the world including begomoviruses which are causing chilli leaf curl virus disease (ChiLCVD). Chilli is known to be affected by 42 viruses. Among them, twenty two of them reported to occur naturally and the rest can infect on artificial inoculation. Chilli leaf curl virus (ChiLCV), chilli leaf curl India Virus (ChiLCINV), tomato leaf curl Joydebpur virus, tomato leaf curl New Delhi Virus (ToLCNDV) are known to be associated with chilli leaf curl diseases (Khan *et al.*,2007).

Recently, it has been studied by Kumar *et al.*,(2015) that the association of five distinct Begomovirus species with six different groups of betasatellites (ChiLCV, Pepper leaf curl Bangladesh Virus (PepLCBV) , Tomato leaf Curl Virus (ToLCV), ToLCNDV, Papaya leaf curl virus (PaLCuV) and beta satellites like ToLCBDB, ChilliCB, ToLCJoB, CroYVMB, RaLC).

There were some several workers who reported that transmission of number of strains of TMV by mechanical sap inoculation of chilli. (Palm,1923; Holmes,1937; Nakata

and Takimoto,1940; Doolittle and Beecher, 1942; Kovachevsky,1940, McKinney,1952; Newton,1954; Miller and Thornberry,1958; Murakshi.1960: Greenleaf *et al.*, 1964; Adsuar *et al.*, 1971; Ragozzino *et al.*, 1972; Bidari,1982). Transmission of Begomovirus is mainly responsible for whiteflies, Brown *et al.*, (2015) described.

2.4. Chilli Leaf Curl Diseases

The chilli leaf curl diseases on chilli was first reported in Pakistan by Shih *et al.* (2003)and in India by Senanayake *et al.* (2006) . But it is most destructive diseases of chilli in India and Bangladesh also. Due to leaf curl diseases of chilli, tropical and subtropical regions of the world where pepper is being cultivated face heavy losses. (Mishra *et al.*; 1963; Dhanaraj and Seth, 1968; Chattopadhyay *et al.*, 2008). If there is severe ChLCV infection, it can cause 100% fruit yield loss. (Kumar *et al.*, 2011). Leaf curl diseases of chilli is currently becoming serious problem in all major chilli growing area of India which accounts for second largest share of chilli production. Chilli leaf curl diseases which is caused by Begomoviruses which are the most catastrophic viruses of chilli plant. One of the major limiting factor for chilli production in the Indian subcontinent is the chilli leaf curl diseases which is occurred by Chilli leaf curl virus. (Briddon *et al.*;2003; Senanayake *et al.*, 2006; Shih *et al.*; 2003).

2.5. Symptomology

Hussain (1932) observed leaf crinkle or leaf curl complex on chilli Leaf curl which is caused by thrips caused by thrips results upward curling of leaves and interveinal buckling. The representative symptoms of the disease compose of leaf curling, leaf rolling, puckering, vein enation, shortening of internodes and petioles and growth retardation (Senanayake *et al.* 2007). Irregular scarping of epidermis can also be noticed. (Johnpulie,1939). The etiology of chili leaf curl virus is quite prevalent in tropical and subtropical areas of the world especially southeast and south Asian countries. *Chili leaf curl virus* is generally monopartite begomovirus consisting of single-stranded positive-sense DNA (ssDNA) sizing ~ 2.7 kb (Senanayake *et al.* 2012).

Downward curling of leaf of chilli is caused by mites, partial suppression of lamina near the petiolar end and a shiny bronze color which is shown on the lower surface of the leaves. Infected plants which are tender and young become brittle narrow and thicker. In India, Srilanka and USA, it is observed that, there were curling of leaf margin, reduction in leaf size, vein clearing in Chilli. (Puttarudraih, 1959). It is observed by Fernando and Peiris (1975) that Chilli Leaf curl virus causes vein clearing on young and old leaves and stunting in most of the cases. In mature fruit, curling of fruit can be seen. White flies (*Bemisia tabaci*) are responsible for transmitting virus and it is further reported that ChLCV does not persist throughout the life span of vector.

Misra *et al* (1963) and Muniyappa and Veeresh (1984) observed that abaxial curling of the leaves accompanied by puckering, thickening and swelling of the veins.

Most prominent symptoms are noticeable on dorsal side such as vein clearing, followed by venial distortion, swelling of veins and veinlets were reported by Muniyappa (1980) and Ravi (1991).

It is found by Dooria and Bindra (1997) that mite density had to be at least 10-15 individual/leaf to produce leaf curl and necrosis, and symptoms were more pronounced on plants receiving 30-50 mites/leaf. After doing treatment with an insecticides, infected plants never recover, if it is aviral infection, contrary to the plants infested by mites and thrips. The economic threshold level has been determined as 1 mite/leaf, was studied by Ukey *et al.* (1999). Thrips in chilli makes the young leaves and buds brittle, resulting in defoliation and total crop loss. Corky tissues are developed in infested fruits. (Seal *et al.*, 2006).

2.6. Identification and Diagnosis of Chilli leaf curl virus

The *chilli leaf curl virus* (ChLCV) was reported first in Pakistan by Shih *et al.*; and in India by Senanayake *et al.*, (2006).

It is also reported that adult and nymphs of *Scirtothrips dorsalis* suck the cell sap of leaves, cause rolling of leaf upward and leaf reduction by Sanap and Nawale (1987).

The appearance of chilli plant is changed by a heavy infestation of *Scirtothrips dorsalis* which is called “chilli leaf curl”. The presence of *Scirtothrips dorsalis* can be identified by the appearance of discolored or disfigured plant parts.

Egyptian isolates of *Beet curly top virus* (BCTV) and TYLCV were inoculated by Abdel-Salam (1990). Not only the Egyptian isolates but also isolates of TYLCV from Jordan respected positively showing their strong serological relationship, when tested serologically using agar gel double-diffusion test with an authentic American BCTV antiserum. These were also confirmed by using immunosorbent electron microscopy test.

It was suggested by Polizzi *et al.*, (1994) depending on the temperature and the time of infection, the types of symptoms varied. However, it was also observed that stunting, reduced leaf and mild chlorosis, having reduced the number of fruits and fruits size were observed.

The viruses which belong to the genus begomovirus consist of a genome made up of either two genomic components, bipartite (known as DNA-A and DNA-B) or a single component of bipartite viruses (Rojas *et al.*; 2005; Stanley *et al.*, 2005).

During the past few years, monopartite begomoviruses and betasatellites associated with chilli leaf curl diseases have been spreading to major chilli growing regions of the Indian subcontinent. There are some viruses which are associated with chilli leaf curl diseases to date in India such as Chilli leaf curl India virus (ChiLCINV), *Chilli leaf curl Vellanad virus* (ChiLCVV), *Tomato leaf curl Joydebpur virus* and *Tomato leaf curl New Delhi Virus* (ToLCNDV) (Khan, *et al.*, 2006; Kumar, *et al.*, 2011, 2012; Senanayake, *et al.*, 2007; Shih, *et al.*, 2007).

Kumar *et al.*; (2015) has studied recently that, the association of five distinct begomovirus species with six different groups of betasatellites [ChiLCV, *Pepper Leaf curl Bangladesh virus* (PepLCBV), *Tomato leaf curl virus* (ToLCV), ToLCNDV, *Papaya leaf curl virus* (PaLcuV) and beta satellites like ToLCBDB, ChiLCB, ToLRnB, ToLJoB, CroYVMB, RaLC].

Further synergistic interaction among different begomoviruses infecting chilli results in breakdown of natural resistance in otherwise resistant chilli plants to one begomovirus (Singh, *et al.*, 2016).

2.7. Chilli leaf curl (ChLC) diseases incidence (%) and diseases severity (%)

Incidence of leaf curl diseases is extent to 40-55% on chilli varieties, viz, IC 374, IC #32, IC 2345, IC 3412 and NP was observed (Mishra *et al.* 1963).

In some district of northern Karnataka, a survey was assess incidence of chilli leaf curl viral complex on chilli. There are some perenial types with small pungent fruits in Tarai region of Uttar Pradesh were shown to be resistant to viruses. Selection from crosses between the perennial local types and NP-46 A was released under the name of Pant C-1 and Pant-2 and they were known to be unsusceptible to leaf curl virus (Mathai *et al.*, 1977).

Various workers have reported about natural occurrence of several viruses on chilli and according to them *Chilli leaf curl virus* (ChLCV), *Cucumber mosaic virus* (CMV) and Chilli vein mottle virus (CVMV). (Green, S.K.1992) are most devastating virus that affect chilli cultivation regarding to incidence and yield loss.

It was roving surveys method which was adopted to know the chilli leaf curl diseases incidence in different parts of Dharwad, Belguam, haveri, Gadag, Bagalkot, Bijapur, Gulbarga, Raichur and Bellary districts during the cropping period both kharif and rabi / summer 2002,2003,2004 and 2005.

The highest diseases incidence (up to 100% of plants during December 2004) was detected in farmer's field in Narwa and Tinwari villages at Jodhpur District Rajasthan (Senanayake *et.al.*, 2007).

2.8. Transmission of chilli leaf curl virus

The Gemini viruses are transmitted by *Bemisia tabaci* which are the largest and most economically significant groups of plant viruses. In case of chilli production, the ability of biotype of *B. Tabaci* to transmit Gemini viruses has adverse effect. (Jeffery *et al.*, 1994). It can be transmitted by mechanically, vector or seed.

The leafcurl or leaf crinkle occuring on chillies was caused by *Bemisia tabaci* (*Bemisia gossypiperda*) and that was reported by Hussan (1932).

Costa and Alves (1950) repoted that there are some aphids like *M. Persicae sulz*, *Macrosiphum solanifolii* Ashm and other undefined aphids were responsible to transmit PYV on chilli.

The virus is transmitted by wedge grafting and white fly (*Bemisia tabaci*) which was studied by Park and Fernando (1938), Mishra *et. al.*, (1963) and Dhanraj and Seth (1968).

The transmission of leaf curl virus by *M. Persicae*, *A. gossypii*, *M. Solanifolii*, *M. Pisi* and *A. Spiraecola*, *A. Craccivora* transmitted the virus in persistence manner which was reported by Laird and Dickson (1963).

Prasada Rao (1976) found that PVMV was transmitted by *A. gossypii*. Gowda and Reddy (1989) noticed that typical strain of poty virus mechanically transmitted and nonpersistently transmitted by aphids, *M. persicae*, *A. fabae* and *A. gossypii*.

It was reported by Bidari and Reddy (1986) reported that TSWV none of aphids transmitted the diseases. Only nymphs of *T. Tabaci* isolated and maintained on onion plants transmitted virus to *C. Annuum cv. California wonder* and *Byadgikaddi* and not by *Scirtothrips dorsalis* as it was reported in groundnut.

Gowda and Reddy (1989) Potato virus Y was transmitted by *M. persicae*, *A. gossypii* and *A. craccivora* at 60,80 and 20 percent respectively and not transmissible by *T. Citricidus* and *R. Maidis*. El-Sanusi *et. al.*, (2009). Premchand.

Rao reported that a number of plant species show natural infection. They further reported that the diseases transmitted by whitefly, *Bemisia tabaci* while it subsists on a wide range on its wild or perennial host plants.

Fereres *et.al.*,(2007). reported that several aphids are responsible for transmitting Potatovirus Y (PVY) which indicates that *M. persicae* was the most efficient vector followed by *A. gossypii* and *Acyethosiphon pisum*, *A. fabae* was also capable of transmitting PVY but less efficiently.

Chilli thrip sected as vectors of tobacco streak virus (TSV) in ground nut crops in India. Recently, in Thailand its role as a vector of three tospoviruses (i.e. *Melon yellow spot virus* (MYSV), *Watermelon silver mottle virus* (WsmoV), and *Capsicum cholorosis virus* (CaCV) in field crops was confirmed by Chiemsombat *et al.*,2008).

2.9. Effect on growth and yield contributing characters by ChLCV

Buds which are aborted and flower which are distorted, shoots grow twisted and fruit may be misshapen and rusted. These same symptoms were observed by Vasudeva and Samraj (1948), Sastry and Singh (1973), Muniyappa (1980), Sakia and Muniyappa (1989).

Chilli leaf curl means abaxial and adaxial curling of leaves that was accompanied by puckering and blistering of interveinal areas and thickening and swelling of veins (Senanayake *et al.*, 2006).

Sastry and Singh (1973) reported that TLCV infected plants produced very few fruits. When infected within 20 days after planting and resulting upto 92.3 % yield loss.

2.10. ChLCV Incidence and Severity with Whitefly

One research conducted by Shimul Das (2020) said, the incidence of leaf curl virus in chilli significantly suppress the plant spacing, planting date as it's vector whitefly abundance over mid (February 1) and late planting (March 1).

A study conducted by M. M. Alam (2016) showed that there were positive correlation between disease incidence, disease severity and whitefly.

2.11. Effect of insecticides on leaf curl virus

Pepper leaf curl virus (PeLCV), transmitted by whitefly is a cosmopolitan plant viral disease hampering a wide range of crops. Chemical elicitors have been used to curb the trend regardless of their setbacks on environment and human kind. Disease management studies were performed by treating three nursery beds separately by spraying with malathion prior to transplanting to the field. Misra *et al.*, (2003) observed that efficacy of this insecticides i.e., Naclfmoa 1.9 EC (fermentation metabolite) at 15, 20 and 25mL ha⁻¹, abamectin (Vertimec) 1.9 EC at 20 mL ha⁻¹, dimethoate (Rogar) 30% at 300 mL ha⁻¹, dicofol (Hilfol) 18.5% EC at 500 mL ha⁻¹ and ethion (Phosmite) 50% EC at 250 mL ha⁻¹ were evaluated in a field at Bhubaneswar. Ethion proved superior in controlling the leaf curl disease of chilli.

Noor (2004) conducted a trail in Jodhpur, Rajasthan for two years to control early season pests of chilli av. RCH-1 (*Scirtothrips dorsalis* and *Polyphagotarsonemus*

latus) using different insecticides, applied through various methods. The insecticides used were phorate, monocrotophos, carbofuron, acephate and methyl demeton applied individually or in combination and found phorate 10 G @ 3 g m⁻² applied as seed treatment followed by monocrotophos (0.05%) root dip before transplanting was highly effective in controlling the pests and leaf curl disease.

It was studied by Susheel *et.al.*, (2010) that management by insecticides, imidacloprid 17.8 SL (0.003%) was most effective than spinosad 48 EC (0.02%), malathion 50 EC (0.05%), acephate 75 SP (0.1%) and methyl-demeton 25EC (0.025%).

2.12. Management of Leaf Curl Virus by using bioagents and natural extract

It was studied by Khalequzzaman *et. al.*, (2018) that that cow urine, cow milk, and neem extract showed comparatively better results against control. So, cow urine, cow milk, and neem leaf extract may be sprayed for controlling vector as well as leaf curl virus and increasing yield of chilli.

One investigation by Sugandhika *et.al.*, (2021) found that sea weed extract (SWE) induces plant immunity but no effect on vector control. Therefore, SWE with recommended insecticides is better to increase the immunity of plants for the effective management of CLCVD.

Zeeshan N. *et. al.*, (2018) showed that Neem seed kernal (NSK) showed the most effective result agninst chilli leaf curl virus disease.

A study was undertaken by Arun kumur, (2011) to determine efficacy of bio-agents like Raw Cow Milk (RCM) and *Trichoderma viride* in managing the disease. Chilli seeds were treated with RCM for 24 hours in 1:1 ratio (i.e. RCM diluted to 50% by adding water) at the room temperature and *T. viride* (6 g Kg⁻¹ seed) and *T. viride* (10 g m⁻²) in nursery soil followed by dipping of nursery-raised saplings in RCM (15%) for 20 minutes before transplantation. Treatment of bio-control agents was found superior over control in all the replications disease.

The plants inoculated with the chitosan based formulation of *Pseudomonas sp.* (206(4) +B-15+ JK-16) recorded the highest activity of ISR molecules and recorded maximum plant height, total biomass, chlorophyll content, fruit number and yield viral load in plants inoculated with both chitosan and *Pseudomonas sp.* 206(4) + B-15+ JK-16 which was studied by Shefali Mishra *et.al.*, (2014).

The capability of Trichoderma bioagents to release salicylic acid under in vitro conditions appear to have prominent role in orchestrating PeLCV suppression (up to 50%). Interestingly, no difference was recorded in the specific activity of the destressing enzymes indicating a likely early ROS accumulation at infection sites leading to restricted virus spread. Overall, Trichoderma could help to counteract viral diseases. *T. harzianum* T8, Atroviridae T2 and Polysporum T1 have the capability to reduced PeLCV in peppers plants was studied by Lanvin Rochal Kepngop Kouokap *et.al.*, (2021),

It was studied by Alvis *et.al.*, (2014) that treatment done by neem leaf extract showed lower disease incidence than other treatments and under this treatment there was the highest yield of Okra.

The management of leaf curl disease, by plant products showed that Neem Seed kernal extract (5%) found most effective than Karanj and Tumba seed extract was studied by Susheel *et.al.*, (2014).

CHAPTER III

MATERIALS AND METHODS

The experiment was conducted during the period from December 2020 to June 2021 to investigate the eco-friendly management of chilli by using different natural extract. In this chapter a brief explanation of the experimental period, location, soil and climate condition of the experimental area and materials & methodology are included.

3.1. Experimental Site

The research was conducted at the Central Farm of Sher-e-Bangla Agricultural University (SAU), Sher-e-Bangla Nagar, Dhaka-1207, Bangladesh. The experiment was carried out during rabi season. The location of the experimental site is situated at 90° 22 ' E longitude and 23° 41' N latitude. The altitude of 9 meters above the sea level. The experimental site is presented in appendix I.

3.2. Agro-ecological region

The experimental field belongs to the agro-ecological region of the Madhupur Tract (AEZ28). The area was under the sub-tropical monsoon climate, which is characterized by heavy rainfall during Kharif season (April to August) and scanty of rainfall during the rest of the year. The landscape comprises level upland, closely or broadly dissected terraces associated with either shallow or broad, deep valleys.

3.3. Characteristics of soil

The soil of the experimental site belongs to the general soil type, Shallow Red Brown Terrace Soils under Tejgaon Series. Top soils were clay loam in texture, olive-gray with common fine to medium distinct dark yellowish mottles. Soil pH ranged from 6.1-6.3 and had organic matter 1.29%. The study area was flat having available irrigation and drainage system and above flood level. Soil samples from 0-15 cm depths were collected from experimental field. The analysis were done by Soil Resource and Development Institute (SRDI), Dhaka. The physicochemical properties of the soil is presented in Appendix II.

3.4. Weather conditions during the experiment

The weather condition of the experimental site was conducted under the sub-tropical monsoon climate. It is characterized by heavy rainfall during Kharif season (April to September) and in the Rabi season (October to March) low rainfall during Rabi season associated with moderately low temperature, low humidity and short day. There was no rainfall during the month of December, January and February whereas, little rain in March. For this, Rabi is the more favourable for vegetable production. Details of the meteorological data in respect of temperature, rainfall and relative humidity during the study period were collected from Bangladesh Meteorological Department, Agargaon, Dhaka-1207 (Appendix III) .

3.5. Planting material used in the experiment

Planting materials were collected from Local farmer of Brahmanbaria, Chittagong, Bangladesh.

3.6. Raising of seedlings

The seedlings of local variety of chilli which is popularly known Haitta Morich in Brahmanbaria were collected from local farmer . The raised the seedling without using any insecticides and pesticides on the died and wet water hyacinth.

3.7. Experimental Treatments

Total ten treatments were used in this experiment. Sterilized distilled water was sprayed with each spray in control plots. First foliar spray was applied at 35 days after transplanting, Second spraying was applied 45 days after transplanting and the third one was applied 55 days after transplanting. Those which plant were treated by only Trichoderma, before planting all plants were dipping with Trichoderma.

A total 10 treatments are stated below

T₀ = Control (Without any treatment)

T₁ = Neem Leaf Extract (0.03% @ 5ml/L)

T₂ = Mehogoni Leaf extract (5% @ 5ml/L)

T₃ = Garlic extract (0.03% @ 5ml/L)

T₄ = 50% Trichoderma + 50% Raw cow milk

T₅= Malathion (.05% @ 1ml/L)

T₆= Lantana leaf extract (0.03%@3ml/L)

T₇ = 50% water+50% raw cow milk

T₈= Trichoderma (0.15% @2ml/L)

T₉ =Marigold leaf extract(.03% @3ml/L)

3.7.1. Preparation of aqueous treatments

Extract of Neem leaf, Garlic cloves, Mehogoni leaf, Lantana leaf, marigold leaf were prepared by weighting 100 gm of blended air dried plant material using electronic balance and then steeped in 1 liter of tap water. The mixture was strain by using cloth to obtain the aqueous extract and 10 ml of mild soap with the mixture were added so that the could attach with leaf surface. 50% Trichoderma was mixed with 50% of water, 50% Trichoderma and 50% raw cow milk were mixed to spray. 1 ml of malathionin was mixed with 1 liter of water for spraying it on the plant. Each of the prepared solution were sprayed by using Knapsack sprayer, 35, 55,75 days after transplanting according to Zeeshan N. *et.al.*,2018 recommmendation. The spraying was done after 3 pm when the sun light was low. It was done to prevent the possibility of the sun rays disintegrating the active ingredients in the aqueous extracts after the application.

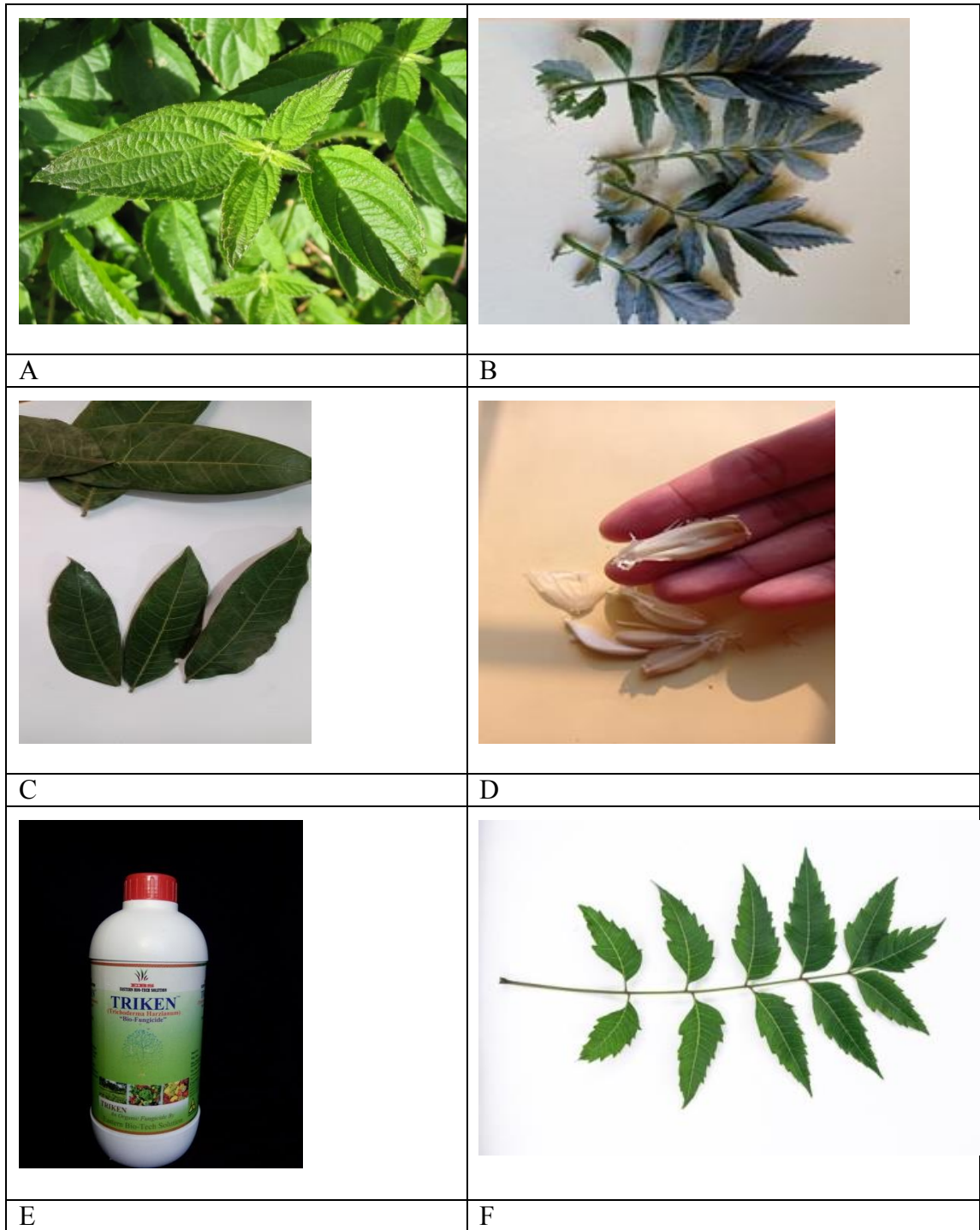


Plate 1: Material used for treatments (A) Lantana leaf, (B) Marigold Leaf, (C) Mahogoni Leaf, (D) Garlic Clove, (E) Tricoderma, (F) Neem Leaf

3.8. Design and layout of the experiment

The factor of the experiment was laid out in a Randomized Complete Block Design (RCBD) with three replications where the experimental area was divided into three blocks representing the replications to minimize the soil heterogeneous effects. The length of the experimental area 20 m and width 10m and the total area of the experimental plot was 200 m² . The total area is divided into three equal blocks. Each block was divided into 10 plots where 10 treatments combination were allotted at random. There were total 30 unit plots in the experiment. The size of the each plot was 2.67 m × 1.67 m. The distance maintained between two blocks and two plots were 0.75 m and 0.4 m, respectively. Side drainage spacing was .5m in each side respectively. Plant to plant distance was 45 cm and row to row distance was 90 cm .

3.9. Land preparation

At the last week of December 2020 the main plot was selected for conducting the experiment was opened with a power tiller, and left exposed to the sun for a week. Then the land was harrowed, ploughed and cross-ploughed several times followed by laddering to obtain good puddle condition. The weeds and different kind of stubbles were removed to make it clean. In accordance to the experimental design and layout, the experimental area was partitioned into different unit plots. Different organic and inorganic fertilizers were mixed with soil.

3.10. Fertilizer and manure application

During final land preparation, fertilizer was applied following the recommendations (Krishi Projukti Hatboi) of Bangladesh Agriculture Research Institute (BARI, 2019) . Before transplanting seedlings to main field from seed bed, well decomposed cow dung (10t/ha) was applied at the time of fine land preparation. The sources of fertilizers used for N,P,K,S and Zn were urea (410 Kg/ha), TSP (300Kg/ha), MP(200Kg/ha), Zinc sulphate (15Kg/ha) respectively. The entire amount of TSP and MP were applied during final land preparation. Only urea was applied in three equal instalments 30, 45 and 60 days after transplanting. Application of potassium as K₂SO₄ will increase the quality of chilli. The used doses of manure and fertilizer are given below in table form

Table 1. Does of fertilizer application

Name of fertilizer and manure	Does/ha
Cowdung	10 ton
Urea	410 kg
TSP	300kg
MP	200kg
Zn	15 kg

Source : Krishi projukti Hat Boi(2019)

3.11. Selected variety Against ChLCV

One local variety called HAITTA MORICH (Local name) which is susceptible to chilli leaf curl disease from Brahmanbaria was considered for this experiment.

3.12. Transplanting of seedlings

Healthy and uniform sized 35 days old seedlings were uprooted separately from water hyacinth. The seedlings were transplanted in the pits of the experimental plots at afternoon on 26th December, 2020 maintaining a spacing 45 cm and 90 cm between the row and the plants respectively. After that, light irrigation was given immediately after transplanting by using water can. In order to gap filling and to check the boarder effect, some extra seedlings were also transplanted around the border area of the experimental field.

3.13.1. Intercultural operations

Various intercultural operations were done for better growth and development of the plants during the period of the experiment.

3.13.2. Gap filling

Gap filling was done in place of dead or injured wilted seedling in the field using healthy seedlings of the same stock previously planted in border area. The transplanted seedlings in the experimental plot were kept under careful observation. To minimize transplanting stock, those seedlings were transplanted with a big mass of

soil with roots . Replacement was done with healthy seedling having a bowl of earth. The transplant were given shading and watering for 7 days for proper establishment.

3.13.3. Weeding and mulching

Whenever necessary to keep crop free from weeds, weeding was conducted and for better soil aeration and to break the soil crust, mulching was done. Four subsequent weeding were done manually at 15,30,45 and 55 DAT to keep the plot free from weeds.

3.13.4. Staking

Staking was given to each plant by bamboo stick to keep them erect, when the plants were well established.

3.13.5. Drainage

Adequate water effectively drained out at the time of heavy rains.

3.13.6. Irrigation

Throughout the growing season, adequate irrigations were given and when necessary. Immediately after transplanting, the plot was semi flooded by irrigation. The crop was irrigated when needed depending on the moisture status of the soil and requirement plants.

3.14. Data Collection

Five plants from each plot were randomly selected for data collection. The collected parameters were shown during field experiment.

- ❖ No of infected plant /plot
- ❖ No. of branches/ plant
- ❖ Plant height(cm)
- ❖ Days to first flowering
- ❖ 50% flowering
- ❖ Length of fruit (cm)
- ❖ Fruit girth (cm)
- ❖ Population of white fly
- ❖ Yield (Kg/ha)

3.14.1. Description of data collection

No of infected plant was counted by visually. Plant height, length of fruit were measured by using wooden scale in centimeter and by using measuring tape fruit grith was measured in centimeter. White fly counting was usually done between 6.00-8.00 a.m. when the environment was cooler and white flies were relatively immobile than later in the day as reported by Fauquet *et.al.* (1987) The total adult white fly population on the five topmost expanded leaves of five plants were determined . Yield of per plot was calculated by the sum of fruit yield of selected five plants.

3.15. Identification of ChLCV by visual observation

Identification of the virus diseases was done by mainly through visual observation of typical symptoms of ChLCV infection like upward curling, cupping, interveinal buckling, crinkle with or without marginal chlorosis, smaller leaflets and stunting of the plant (Green and Kaloo,1994 and Sinistera *et al.*, 2000). The incidence of ChLCV was calculated by counting their infected plants at 35, 55 and 75 DAT on the basis of the appearance of symptoms. The plants were inspected every morning to note the appearance of symptoms starting from the following day of transplantation.



Plate 2: Infected leaf and healthy leaf

3.16. Diseases Incidence (%)

By using direct counting method, chilli leaf curl disease incidence was recorded. Infected plants were counted at regular intervals and the final percentage worked out. The following formulas were used to calculate the percentage of disease incidence and severity-

Disease incidence (%) was computed by formula developed by (Mayee and Datar, 1986)

$$\text{Disease incidence (\%)} = \frac{\text{No.of plants infected}}{\text{Total number of plant}} \times 100$$

3.17. Percent Disease Index

Percept diseases index was calculated by the formula given by Wheeler (1969)

$$\text{Percept Disease Index} = \frac{\text{Total disease rating}}{\text{Total no of observation X Maximum Scale}} \times 100$$

Scale	Description	Disease Reduction
0	No symptoms on plants	Immune(I)
1	1% or less leaves exhibiting symptoms	Resistant(R)
3	1 to 10 % leaves exhibiting symptoms	Moderately Resistant (MR)
5	11 to20 %leaves exhibiting symptoms	Moderately Susceptible (MS)
7	21 to50 %leaves exhibiting symptoms	Susceptible(S)
9	21 to50 %leaves exhibiting symptoms	Highly Susceptible(HS)

3.18. Harvesting and post harvest operation

The harvest of crop was completed at 75 DAT. The crop was harvested plot wise. The harvested fruits were sorted into individual bags for each plot separated by healthy and infected . They were taken to the khamar shed after sorting the fruit on the floor, weighed separately.

3.19. Yield increase (%)

Percept yield reduction in terms of weights was calculated as follows (Mousanejad *et al.*, 2010).

$$\text{Yield increase (\%)} = \frac{\text{Yield of treated plot} - \text{Yield of untreated plot}}{\text{yield of untreated plot}} \times 100$$

3.20. Statistical Analysis

The collected data were compiled and analysed statistically using the analysis of variance technique with the help of a computer package program Statistics-10 and the mean difference were adjusted by Least Significance Difference (LSD) test at 5% level of probability (Gomez and Gomez,1984). The means of growth and yield data were compared by LSD Bar Diagram and graphs were also used to interpret the data as and when necessary.

Chapter IV

RESULTS AND DISCUSSION

The present experiment was conducted to study the management of leaf curl disease of chilli under natural condition. The data on diseases incidence, percent disease index, white fly population, percent disease reduction, growth, yield and yield attributing characteristics were studied. To full fill the objectives of the results of the research were described under the following headings and subheadings.

4.1. Symptomology

The visible symptoms observed were downward curling of leaves, light and vein clearing, puckering of leaf lets, stunting and bushy appearance because of the reduction of internodal length to complete sterility and infected twig almost shows upward or downward of curling leaves with aborted flower and misshapen fruit.



Plate 3 : ChLCV infected leaf, flower and fruit

4.2. Analysis of Variance for the characters observed

The analysis of variance for the characters observed in different treatment at 75 DAT. The analysis of variance indicated the presence of significant variation among the treatments for the characters are presented in table 2.

Table 2. Analysis of variance for the characters observed at 75 DAT

Sl. No.	Characters	Mean Sum of Sum Squares	
		Treatments	Error
1	Plant height	308.54**	22.08
2	No. of branches per plant	11.86**	1.58
3	Days to first flowering	631.11**	.078
4	Days of 50% of flowering	624.504**	0.248
5	Length of fruit	15.85**	6.33

** Significant at 1% level

4.3. Effect of different treatments on chilli leaf curl disease incidence (%) at 35, 55, 75 DAT

There were significant variations found in different treatments on percent disease incidence against Chilli leaf curl disease on chilli. The result of chilli leaf curl disease incidence are shown in table 3.

The diseases incidence of ChLCV among different treatment was varied significantly. The disease incidence (%) of chilli was ranged from 4.97 to 16.99, 7.82 to 18.41 and 9.97 to 20.97 at 35,55, 75 DAT, respectively .

At 35 DAT , the lowest (7.03%) disease incidence was found in T5 (Malathion @3ml/L) followed by T8 (Trichoderma, 7.03) in T1 (Neem leaf extract, 7.24), T3 (Garlic extract,8.067) which were statistically similar. While the highest (16.99%) disease incidence was found in T0 (Control) preceded by T9 (Marigold leaf extract) was 14.45.

At 55 DAT, the lowest (7.82%) disease incidence was found in T5 (Malathion) and the highest disease incidence(18.41%) was in T0(Control). The percent disease

incidence of T₁ (Neem leaf extract), T₅ (Malathion) and T₈ (Trichoderma) were 8.893, 8.51 and 7.82 respectively which were statistically indistinguishable.

At 75 DAT, statistically more variation was found in disease incidence (%) of ChLCV. The lowest (9.97%) ChLCV disease incidence was found in T₅ (Malathion) and the highest (20.97%) disease incidence was in T₀ (Control).

Table 3. Effect of different treatment on Disease Incidence (%) of ChLCV at 35,55, 75 DAT

Treatment	Disease Incidence (%)		
	35 DAT	55 DAT	75 DAT
T ₁	7.24 f	8.89 f	11.30 fg
T ₂	11.10 de	12.71d	16.12 bcd
T ₃	8.06 f	10.75 d	13.15 ef
T ₄	10.17 e	12.74 d	14.47 de
T ₅	4.95 g	7.82 f	9.97 g
T ₆	12.20 cd	14.03 cd	15.81 cd
T ₇	12.52c	8.51 c	16.82 bc
T ₈	7.03 f	16.14 f	10.07 g
T ₉	14.45b	18.41 f	18.12 b
T ₀	16.99 a	18.41a	20.97 a
CV(%)	5.74	7.20	10.683

T₁ (Neem leaf extract), T₂ (Mahogoni leaf extract), T₃ (Garlic extract), T₄ (Trichoderma + Raw cow milk), T₅ (Malathion), T₆ (Lantana), T₇ (Raw milk), T₈ (Trichoderma), T₉ (Marigold leaf extract) DAT = Days after transplanting, CV = Coefficient of variance.

4.4. Effect of different treatment on percent disease index of ChLCV at 35, 55, 75 DAT

The percent disease index of ChLCV was varied on the basis of different treatment significantly. It was found that the percent disease index of ChLCV was ranged from 3.77(%) to 15.26(%), 6.38 to 16.41(%) and 8.82(%) to 19.25(%) at 35,55,75 DAT respectively. The results of percent disease index are present in table 3

The lowest (3.77) percent disease index was found in T₅ (Malathion) which was moderately resistant and the highest (15.26) was in T₀ (Control) which was moderately susceptible according to disease rating scale at 35 DAT.

The lowest (7.08) percent disease index was found in T₈ (Trichoderma) and the highest (25.13) was in T₀ (Control) at 55 DAT where T₈ (Trichoderma) was

moderately resistant to ChLCV and the rest one was susceptible according to disease rating scale.

At 75 DAT, the lowest (8.68) percent disease index was in T₈ (Trichoderma) was moderately susceptible according disease rating scale and the highest(28.99) was in T₀ (control).

Table 4. Effect of different treatment on percent disease index (PDI) of ChLCV in chilli at 35, 55, 75 DAT

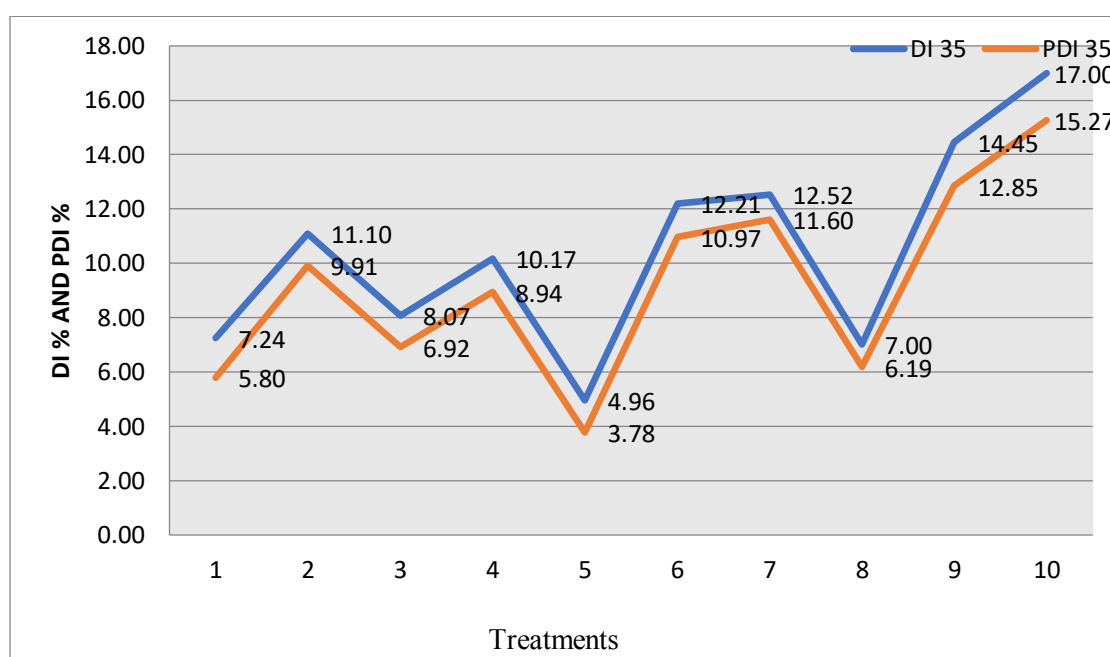
Treatment	35 DAT		55 DAT		75 DAT	
	Percent disease index	Reaction	Percent disease index	Reaction	Percent disease index	Reaction
T ₁	5.79	MR	7.26	MR	9.52	MR
T ₂	9.09	MR	11.59	MS	14.59	MS
T ₃	6.92	MR	9.54	MR	11.56	MS
T ₄	8.94	MR	11.76	MS	13.51	MS
T ₅	3.77	MR	6.38	MR	8.82	MR
T ₆	10.96	MS	13.58	MS	19.54	MS
T ₇	11.60	MS	13.22	MS	15.55	MS
T ₈	6.18	MR	7.08	MR	8.68	MS
T ₉	12.84	MS	14.63	MR	17.54	MS
T ₀	19.26	MS	25.13	S	28.99	S

T₁ (Neem leaf extract), T₂ (Mahogoni leaf extract), T₃ (Garlic extract), T₄ (Trichoderma + Raw cow milk), T₅ (Malathion), T₆ (Lantana), T₇ (Raw milk), T₈ = Trichoderma, T₉ = (Marigold leaf extract)* MR = Moderately Resistant, MS = Moderately Susceptible, S = Susceptible

4.5. Performance of different treatments against ChLC disease incidence (%) and percent disease index at 35, 55, 75 DAT

4.5.1 Performance at 35 DAT

Among all the tested treatments, the lowest (4.95%) disease incidence was in T₅ (Malathion) and the lowest percent disease incidence was 3.77 on the other hand, the highest (19.26) percent disease index and the highest (16.99) disease incidence was in T₀ are presented in Fig.1 .

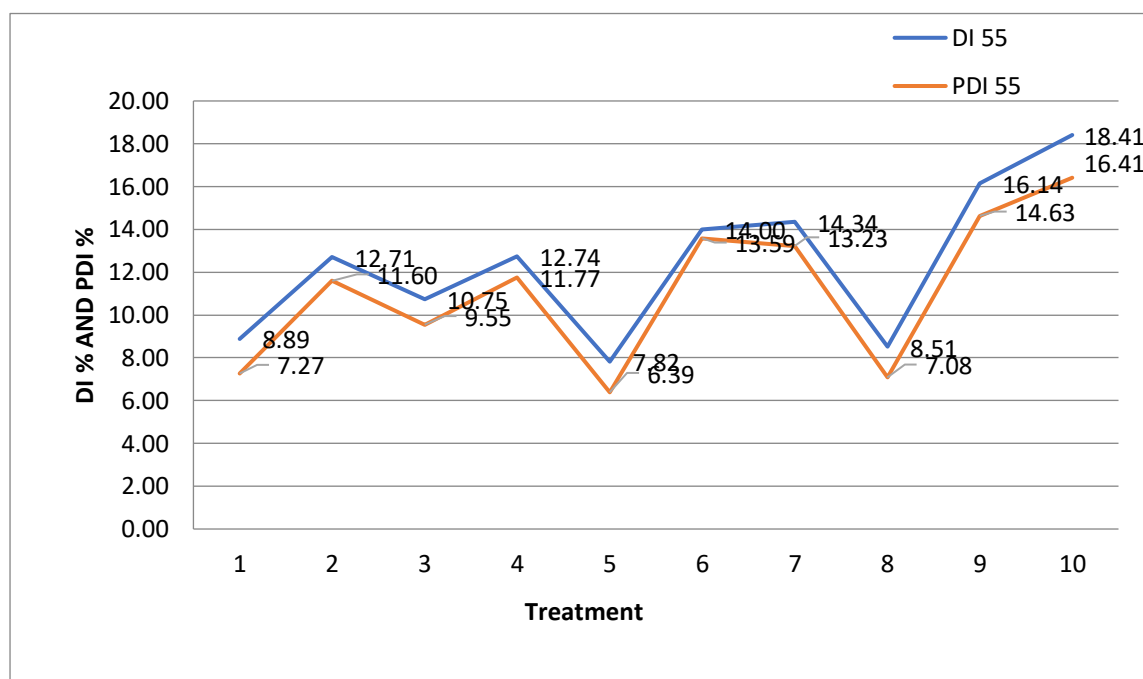


T₁ (Neem leaf extract), T₂ (Mahogoni leaf extract), T₃ (Garlic extract), T₄ (Trichoderma + Raw cow milk), T₅(Malathion) , T₆(Lantana) , T₇ (Raw milk) T₈= Trichoderma , T₉ = (Marigold leaf extract)

Figure 1: Performance of different treatments percent disease incidence and percent disease index at 35 DAT

4.5.2 Performance At 55 DAT

The lowest (7.82%) disease incidence and the lowest (6.38%) percent disease incidence were in T₅ (Malathion) followed by the disease incidence and percent disease incidence was 8.51 and 7.8 respectively in T₈ (Trichoderma). On the other hand, the highest percent disease incidence was 18.41 and the highest percent disease index was 16.41 in T₀(Control),are presented in figure 2.



T₁ (Neem leaf extract), T₂ (Mahogoni leaf extract), T₃ (Garlic extract), T₄ (Trichoderma + Raw cow milk), T₅(Malathion) , T₆(Lantana) , T₇(Raw milk) T₈=Trichoderma , T₉ = (Marigold leaf extract)

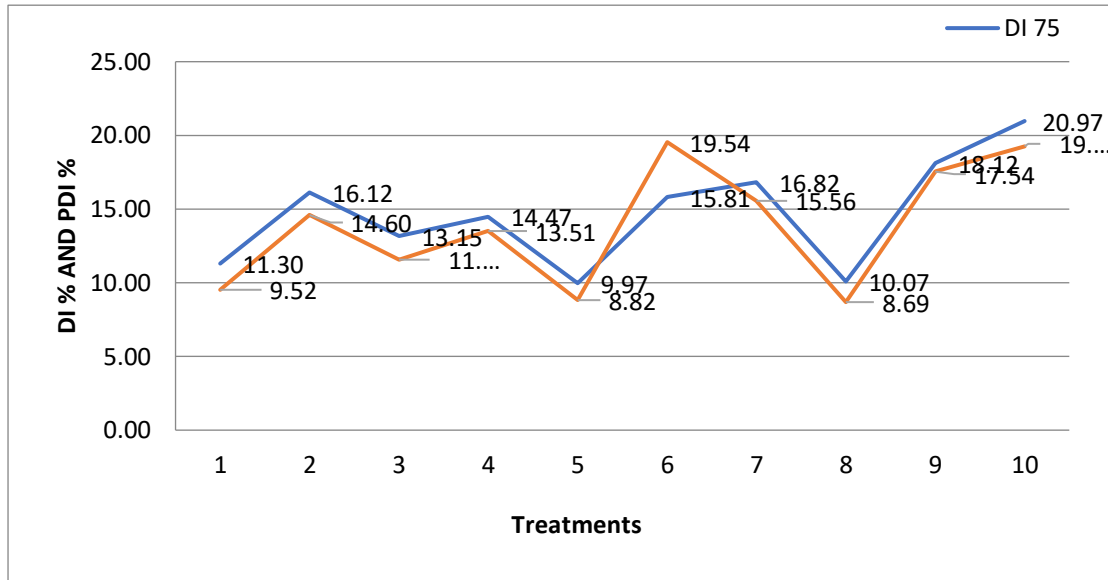
Figure 2 : Performance of different treatments on percent disease incidence and percent disease index at 55 DAT

4.5.3. Performance at 75 DAT

Among all the tested treatments, the lowest (9.97%) disease incidence and the lowest (8.82) percent disease incidence was in T₅ (Malathion) followed by the disease incidence and percent disease incidence was 17.39% and 8.68 respectively in T₈ (Trichoderma).

On the other hand, the highest disease incidence and percent disease index was 20.97 and 28.99 respectively in T₀(control) followed by 18.12% disease incidence and

17.45 percent disease incidence was in T₉ (Marigold leaf extract). Results are showed in figure 3 .



T₁ (Neem leaf extract), T₂ (Mahogoni leaf extract), T₃ (Garlic extract), T₄ (Trichoderma + Raw cow milk), T₅ (Malathion) , T₆ (Lantana) , T₇ (Raw milk) T₈ = Trichoderma , T₉ = (Marigold leaf extract)

Figure 3. Performance of different treatments on percent disease incidence and percent at 75 DAT

4.6. Effect of *Chilli Leaf Curl Virus* on growth attributing characters among different treatments at 75 DAT

There are appreciable variation found in number of infected leaves, plant height, number of branches against *Chilli leaf curl virus*. These result are presented in table 3.

4.6.1. Number of branches/ plant

At 75 DAT, the highest (12.33) number of branches were in T₄ (Trichoderma + raw cow milk) and followed by the highest (12.00) number was in T₃ (Garlic extract) . The lowest (9.00) number of branches was in T₀ (Control) .

4.6.2. Plant Height (cm)

At 75 DAT, it was found that in T₅ (Malathion), there was highest (55.44cm) plant height and then the highest (44.63 cm) plant height was in T₈ (Trichoderma). The lowest (25.90cm) plant height was in T₀ (Control).

4.6.3. Infected leaves

At 75 DAT, the highest (94.66) number of infected leaves was in T₀ (Control) and the lowest (7.90) was in T₅ (Malathion). T₃ (Garlic extract) and T₄ (Trichoderma + Row cow milk) were respectively 12.90 and 15.12 which were statistically identical.

Table 5 : Effect of Chilli Leaf Curl Virus on growth attributing characters among different treatments at 75 DAT

Treatment	Plant height(cm)	No. of branches/plant	No. of infected leaf /plant
T ₁	40.86 bc	11.00	8.67 f
T ₂	49.22 c	12.33b	17.09 e
T ₃	42.10 ab	12.00 b	15.12 de
T ₄	42.09 bc	12.33 b	12.90 de
T ₅	55.44 a	9.67 a	7.90 f
T ₆	32.41 d	9.33 cd	26.88 d
T ₇	29.22 d	9.33 cd	44.33 c
T ₈	44.66 bc	12.33 b	25.03 d
T ₉	25.42 d	8.66 d	53.09b
T ₀ (control)	25.90 d	9.00 cd	94.66 a
CV (%)	12.12	11.25	14.94

T₁ (Neem leaf extract), T₂ (Mahogoni leaf extract), T₃ (Garlic extract), T₄ (Trichoderma + Raw cow milk), T₅ (Malathion), T₆ (Lantana), T₇ (Raw milk) T₈ = Trichoderma, T₉ = (Marigold leaf extract) DAT = Days after transplanting , CV = Coefficient of variance

4.7. Effect of different treatments on yield attributing parameters of chilli

Due to applying different treatments on chilli plant, they exposed their yield attributing parameter which are presented in table 6.

4.7.1. Days to first flowering

There were variation under different treatment of days to first flowering. The highest number of first flowering were 99.33 in T₅ (Malathion) and then 95 in T₁(Neem leaf extract).

4.7.2. Fifty percent of Flowering

A significant variation was shown in days to fifty percent of flowering under different treatments. The highest (104.67) number days of fifty percent of flowering was in T₄ (Malathion) followed by 101 was in T₁ (Neem leaf extract). The lowest (55.33) was in T₀ (Control) and then the lowest (66.33) was shown in T₉ (Marigold leaf extract).

4.7.3. Fruit length (cm)

There were a significant variance was found in fruit length under different treatments. The highest (11.31cm) length was shown in T₈ (Trichoderma) followed by 9.44 cm in T₅ (Malathion). The lowest (5.88 cm) fruit length was in T₀ (Control) preceded by (5.89 cm) in T₂ (Neem leaf extract) and both are identically similar.

4.7.4. Fruit grith (cm)

A significant variation was found in fruit grith while applying different treatment. The highest (4.66cm) fruit grith was found in T₅(Malathion) followed by (3.95 cm) in T₇ (Raw cow milk). The lowest (2.67 cm) was in T₀(Control) preceded by (2.75cm) in T₄ (Trichoderma+Row cow milk). All result are presented in Table 6.

Table.6. Effect of different treatment on yield attributing characters

Treatment	Days to 1st flowering	50% flowering	Fruit length (cm)	Fruit grith (cm)
T ₁	86.00 b	91.67 b	6.48 b	3.05 bcd
T ₂	92.00 e	98.00 e	5.89 b	3.55 abcd
T ₃	89.33 c	95.67 c	5.96 b	2.95 bcd
T ₄	99.33 d	104.67 d	7.96 ab	2.75 cd
T ₅	83.33 a	89.33 a	9.44 ab	4.66 a
T ₆	75.33 f	81.33 f	8.01 ab	2.81 cd
T ₇	87.33 g	93.33 g	7.77 ab	3.95 ab
T ₈	60.33 e	66.33 e	11.31 a	3.80 abc
T ₉	55.33 i	61.33 i	8.55 ab	2.97 bcd
T ₀	95.00 j	101.00 j	5.88 ab	2.67 d
CV(%)	0.34	0.56	4.52	1.01

T₁ (Neem leaf extract), T₂ (Mahogoni leaf extract), T₃ (Garlic extract) , T₄ (Trichoderma + Raw cow milk), T₅(Malathion) , T₆(Lantana) , T₇ (Raw milk)T₈ = Trichoderma , T₉ =(Marigold leaf extract) DAT = Days after transplanting , CV = Coefficient of variance.

4.6.3. Effect of different treatments on yield and yield increase over control due to ChLCV disease in chilli

There are significantly variation was found in yield under different treatment. The highest (69.03 kg/ha) amount of yield was in T₅ (Malathion) followed by (65.8kg/ha) in T₁(Neem leaf extract) and the lowest (49.41 kg/ha) was in T₀ (Control). Preceded by (51.19 Kg/ha) in T₉ (Marigold leaf extract). While the highest (39.69 Kg/ha) reduction was in T₄(Trichoderma + Raw cow milk) and the lowest (3.59 t/ha) was in T₉(Marigold leaf extract).

Table 7. Effect of different treatments on yield and yield increase over control due to ChLCV

Treatment	Yield(Kg/ha)	Yield increase over control(Kg/ha)
T₁	65.8 b	32.36 b
T₂	63.14 c	27.01 c
T₃	58.13 f	16.93 e
T₄	59.52 d	18.98 e
T₅	69.03 a	39.69 a
T₆	55.37 g	16.11 e
T₇	52.32 h	5.88 f
T₈	60.66 e	22.78 d
T₉	51.19 i	3.588 f
T₀	49.41	-
CV (%)	1.00	9.89

T₁(Neem leaf extract), T₂(Mahogoni leaf extract), T₃(Garlic extract), T₄(Trichoderma + Raw cow milk), T₅(Malathion), T₆(Lantana), T₇(Raw milk) T₈= Trichoderma, T₉ = (Marigold leaf extract), T₀= control CV = Coefficient of variance.

4.7. No of white fly association per treatment in 35, 55, 75 DAT

There were a significant difference while applying different treatment to control ChLCV under field condition. Shown in Table 8

At 35 DAT, the highest white fly population was 21.00 in T₀ (Control) and the lowest (6.00) was in T₅ (Malathion). In T₁ (Neem leaf extract) and in T₈ (Trichoderma) the population of whitefly were 11.33 and 12.00 are statistically identical. The lowest (21.00) was in T₀ (Control) preceded by T₉(Marigold leaf extract) was 19.33. At 55 DAT, the highest (23.66) population was in T₀(Control) and the lowest (8.67) was in T₅ (Malathion).

At 75 DAT, it was seen that, the highest (27.33) population was in T₀ (Control) and the lowest (11.67) was in T₅ (Malathion).

Table 8. Effect of different treatment on white fly population at 35, 55, 75 DAT

Treatment	Population of whitefly		
	35 DAT	55 DAT	75 DAT
T ₁	11.33 de	14.33 de	17.33 de
T ₂	17.00 bc	20 bc	22.66 bc
T ₃	10.66 e	13.33 e	16.00 e
T ₄	14.33 cd	17.00 cd	20.00 cd
T ₅	6.00 f	8.67 f	11.67 f
T ₆	16.00 bc	19.00 bc	22.66 bc
T ₇	18.00 ab	21.00 ab	24.00 ab
T ₈	12.00 de	15.00 de	17.33 de
T ₉	19.33 a	22.00 ab	24.33 ab
T ₀	21.00 a	23.66 a	27.33 a
CV(%)	13.36	11.24	9.71

T₁ (Neem leaf extract), T₂ (Mahogoni leaf extract), T₃ (Garlic extract), T₄ (Trichoderma + Raw cow milk), T₅ (Malathion), T₆ (Lantana), T₇ (Raw milk), T₈ = Trichoderma, T₉ = (Marigold leaf extract), T₀ = control CV = Coefficient of variance

4.8. Relationship between the white fly and disease incidence at 35, 55, 75 DAT

The relationship between white fly and disease incidence of ChLCV in the field condition are shown in figure 5. The figure is showing a strong positive correlation between the vector of ChLCV of chilli and disease incidence. With the increase of whitefly, disease incidence is also increased. A regression line was fitted between white fly and disease incidence (%) of ChLCV. There are positive correlation between them.

4.8.1 Relationship at 35 DAT

The value of R was 0.927 which has not significant relationship with disease incidence and white fly association.

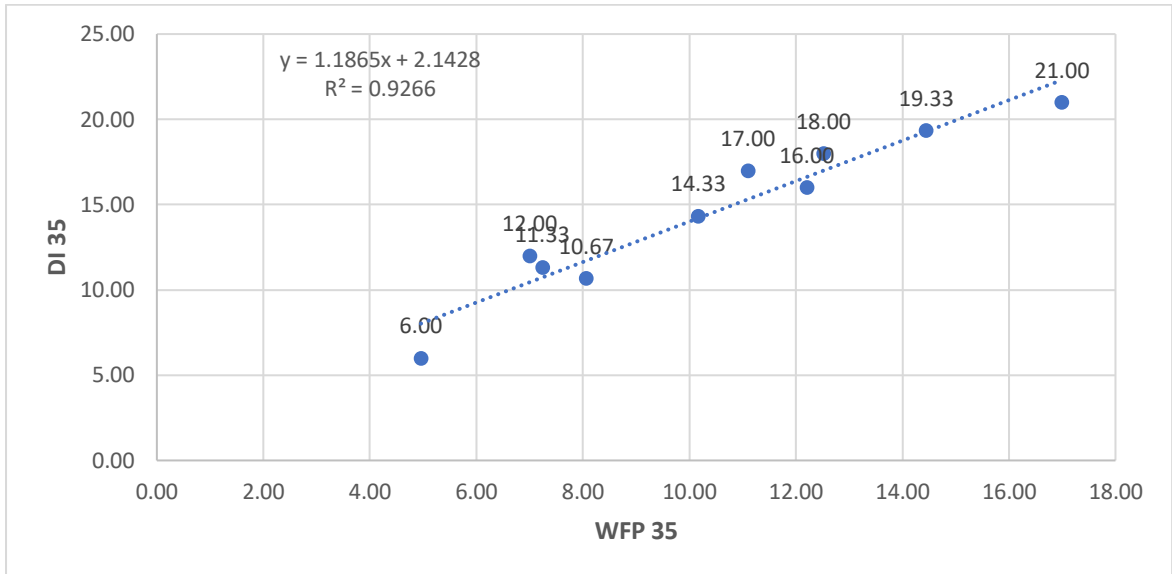


Figure 4 : Relationship Disease incidence between white fly association at 35 DAT

4.8.2 Relationship at 55 DAT

There was no significant relationship with white fly and disease incidence where the value of R was 0.845 .

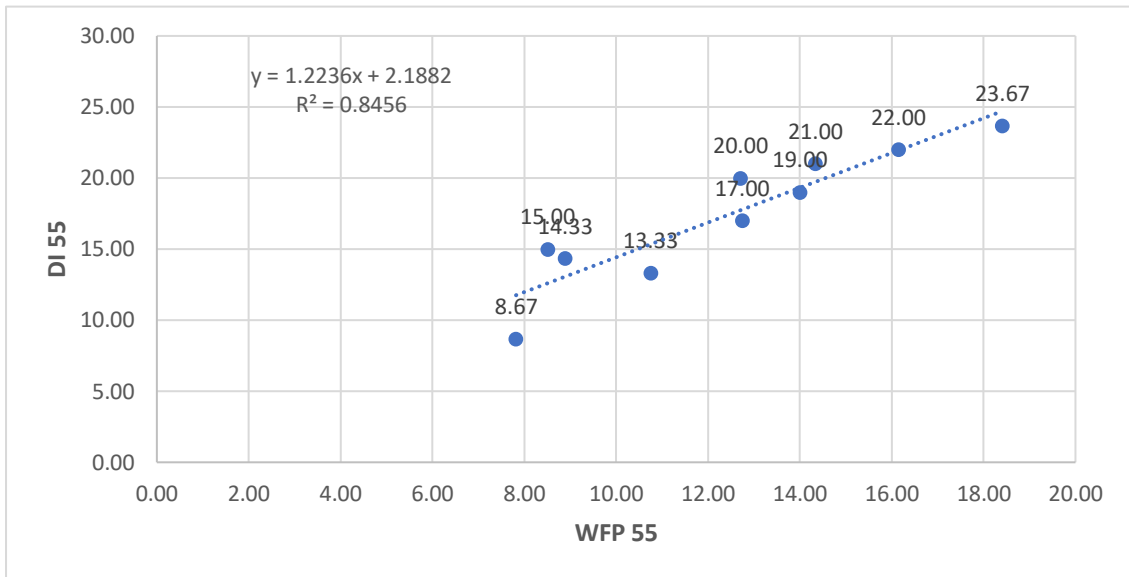


Figure 5: Relationship Disease incidence between whitefly popularion at 55 DAT

4.8.3 At 75 DAT

There is no significant relationship with white fly association and disease incidence where the R value is 0.87

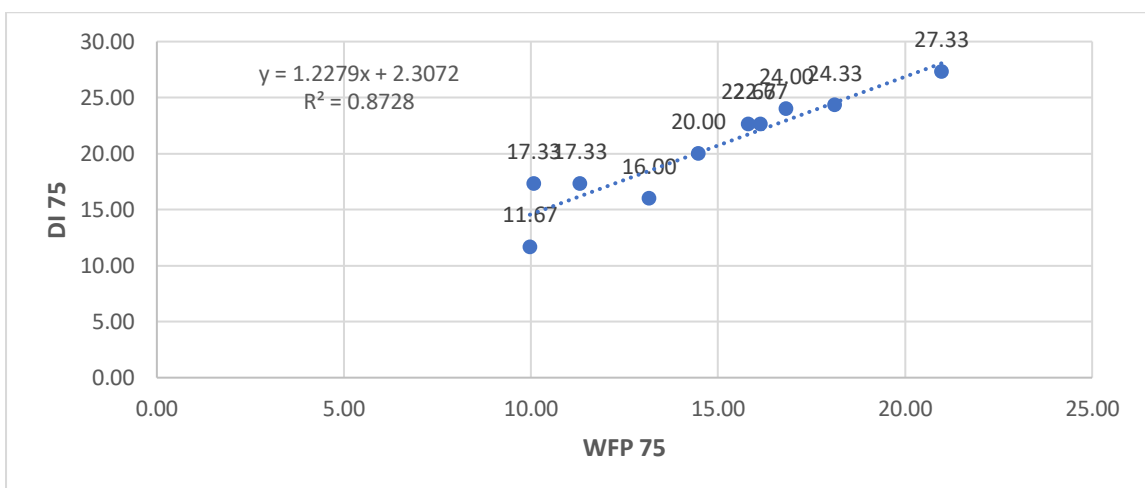


Figure 6 : Relationship between Disease incidence and white fly asocation

DISCUSSION

One of the most important diseases of chilli is *Chilli leaf curl virus* in Bangladesh. It is a threat to chilli cultivation as it is existing in the country for several decades. In Bangladesh, chilli is grown in an area of about 66,235 hectare and its annual production is about 52,215 metric tons which is very low as compared to that of other chilli growing countries in the world and the average yield of green chilli is 1.3 t/ha (BBS, 2018). Chilli leaf curl virus is considered one of the major constrain for the lower yield of chilli in our country.

The present experiment was under taken to study the performance of different treatments of chilli against *chilli leaf curl virus* under field condition. The main objective of this study is to assess the disease incidence (%) and percent disease index of chilli leaf curl virus among different treatments. And to evaluate different natural products, bioagent, insecticides on vector activities to reduce chilli leaf curl disease.

Symptomology

The symptoms observed are downward curling of leaves, light and vein clearing, puckering of leaf lets, stunting and bushy appearance because of the reduction of internodal length with partial to complete sterility and infected twig almost shows upward or downward of curling leaves with aborted flower and misshapen fruit. Similar symptoms were observed by Vasudeva and Samraj(1948); Sastry and Singh(1973); Muniyappa(1980), Sakia and Muniyappa(1989) chilli leaf curl virus is most destructive and chilli leaf curl disease complex means abaxial and adaxial curling of leaves accompanied by puckering and blistering of interveinal areas and thickening and swelling of veins, reported by Kumar, R.V., Singh, A.k and Chakraborty, S.(2012) in different chilli varieties due to ChLCV.

Effect of different treatments on chilli leaf curl disease incidence at 35, 55,75 DAT

In present investigation, it was noticed that the disease incidence(%) due to Chilli leaf curl virus was found almost all the plots at 75 DAT. At 35 DAT, the lowest (7.03) percent disease incidence was found in T₅ (Malathion) and the highest (16.99) percent disease incidence was found in T₀ (control). At 55 DAT, the highest (18.41%)

disease incidence was in T₀ (Control) and the lowest was in T₅ (Malathion). At 75 DAT, the lowest (9.97) percent disease incidence was in T₅ (Malathion) and the highest (20.97) was in T₀ (Control). The highest disease incidence (up to 100% of plants during December 2004) was detected in farmer's field in Narwa and Tinwari villages at Jodhpur District Rajasthan (Senanayake et al. 2007). Zeesan N. et al., 2018 found that neem botanicals showed the lowest (23.45) percent disease incidence under field condition.

Effect of different treatment on percent disease index of ChLCV at 35, 55, 75 DAT

The lowest (3.77) percent disease index was found in T₅ (Malathion) which was moderately resistant and the highest (15.26) was in T₀ (Control) which was moderately susceptible at 35 DAT. The lowest (7.08) percent disease index was found in T₈ (Trichoderma) and the highest (25.13) was in T₀ (Control) at 55 DAT where T₈ (Trichoderma) was moderately resistant to ChLCV and the rest one was susceptible. At 75 DAT, the lowest (8.68) percent disease index was in T₈ (Trichoderma) which was moderately susceptible and the highest (28.99) was in T₀ (Control) which was susceptible to ChLCV. Elvis et al. (2014) found that garlic extract and neem leaf extract showed better result than other treatments.

Relationship between disease incidence (%) and percent disease incidence

The highest (20.97%) disease incidence was recorded in T₀ (Control) 75 DAT and the lowest (9.97%) disease incidence was found in T₅ (Malathion). At 55 DAT, T₀ (Control) showed the highest (18.41) percent disease incidence and T₅ (Malathion) showed the lowest (7.82) percent disease incidence. At 35 DAT, in T₀ (Control), the highest (16.99) percent disease incidence was observed and the lowest (7.03) was in T₅ (Malathion). On the other hand, the highest (15.26) percent disease index was in T₀ (Control) which was susceptible to ChLCV and the lowest (3.77) was in T₄ (Malathion) which is moderately resistant at 35 DAT. At 55 DAT, there were higher (16.41) percent disease index in T₀ (Control) than other treatments while the lower (6.38) was in T₅ (Malathion). At 75 DAT, the highest (19.54) and the lowest (8.20) percent disease index were in T₆ (Lantana) and T₄ (Malathion) respectively. Zeesan N.

(2018) *et.al.*, reported that neem products showed better result where disease incidence was lower (23.45%) than other natural products .

Effect of Chilli Leaf Curl Virus on growth promoting characters among different treatments

The lowest number healthy leaves was found in T₀ (Control) . Significant variation was found in number of branches/ plant ,plant height under different treatment affected by chilli leaf curl virus . At DAT 75,the highest(47.63 cm) plant height was in T₅ (Malathion) and it was said by Mahnaz Nasrabadi *et. al.*,(2021) that there were significant effect of malathion on plant height . Smitha (2002) that the influence of neem product on the plant height was better than other alternatives. The lowest plant height was in T₀ (control) at 35 DAT , 55DAT,75 DAT when it is compared to all other treatment. The number of branching was highest in T₈ (Trichoderma) at 35 DAT, 55 DAT, 75 DAT while the lowest number of branching was in T₀ (Control). During Rabi season, maximum plant height(38.69 cm), no of branches(5.30) per plant were recorded in treatment done by Neem seed kernel which was studied by Zeeshan N *et.al.*, (2018) in Rachi , Jharkhand

Effect of ChLCV on yield attributing parameters of chilli under different treatments

The yield of chilli depends on the number flowers and yield per plant. The lowest (55.33) number of first flowering was in T₀ (Control) and the highest(99.33) was in T₄(Malathion).The highest(69.03 kg/t) yield was in T₅(Malathion), followed by the highest (65.81 kg/ha) was in T₁(Neem leaf extract) and the lowest(49.41 kg/t) was in T₀ . The highest disease incidence of ChLCV causes the highest yield loss of chilli found by Zeeshan(2018). The highest (11.31cm) length was shown in T₈(Trichoderma) followed by 9.44 cm in T₅(Malathion) . The lowest (5.88 cm) fruit length was in T₀ (Control).The highest (4.66cm) fruit girth was found in T₅(Malathion) and the lowest (2.67 cm) was in T₀(Control).The highest fruit yield was in 64.45 kg/ ha under the spraying of Neem seed kernel 5% @ 5ml/L during Rabi season 2015-201 and maximum disease incidence (35.45%) in control with lowest(48 t/ha) yield found by Zeeshan N. *et.al.*,(2018) . One experiment was conducted by Khalequzzaman *et. al.*,(2018) in the field laboratory of Spices Research Center, BARI, Shibganj, Bogra, Bangladesh during Rabi season of 2014-15 to find out the

effect of biopesticides in controlling vector as well as *leaf curl virus* of chilli. The maximum infected plants (26.85%) were recorded in untreated control and the minimum infected plants (14.75%) were in Malathion (0.2%) which was statistically identical to all other biopesticides except cow urine (1:2). The highest yield (18.33 t/ha) was recorded in Malathion (0.2%) and the lowest (11.28 t/ha) was recorded in untreated control. All the biopesticides resulted more yield compared to control.

Research done by Zeesan N *et.al.*,(2018) found that Neem seed kernal showed the highest yield(33.82q/ha) over control

No of whitefly association per treatment at 35, 55,75 DAT

There were significant variance of white fly population after applying different treatments at 35,55,75 DAT . At 35 DAT, the highest(21) number of population was found in T₀ (Control) and the lowest was in T₅ (Malathion) followed by the lowest (10.66) in T₃ (Garlic extract) . At 55 DAT, the highest number of white fly population was in T₀(23.66) and the lowest (8.66) was in T₅ (Malathion) . At 75 DAT , the highest (27.33)population of whitefly was in T₀(Control) and the lowest (11.66) was in T₅ . Malathion showed the lowest number of whitefly studied by Khalequzzaman *etal.*, (2018)

Relationship between White fly association and disease incidence

Under control condition in T₀ , there was highest disease incidence and white fly number . If there is highest white fly population, the disease will be increased. On the other hand, number of whitefly was lowest in T₄(Malathion) and then in T₁ (neem leaf extract) . It was studied by Elvis Asare *et.,al.*(2014) in Ghana that the plant extract significantly reduced the disease incidence the number of white fly was positively correlated with disease incidence. Leaves extracts of four plant extracts i.e. *Calotropis gigantea*, *Zingiber officinale*, *Allium cepa* and *Azadirachta indica* were evaluated at 3 % concentration against eggs hatchability and adult emergence of the whitefly in lab condition. Two consecutive sprays were applied to assess the relative impact of different plant extracts against adult whitefly population and the disease incidence of cotton leaf curl virus disease in filed conditions. Neem leaf was found most effective to inhibit the egg hatchability and adult emergence as compared to other plant extracts applied in lab conditions. In field conditions, it was found most effective to minimize whitefly population and to lower the disease incidence. The leaf

extract of neem leaf at 3% concentration may be used to minimize the whitefly population and to lower the disease incidence of cotton leaf curl virus disease under field conditions observed by Waseem Abbas(2020) . It was studied by Mallikarjuna Rao(1999) *et.al* that the effectiveness of neem product against whitefly .

CHAPTER V

SUMMARY AND CONCLUSION

The study was to evaluate the performance of different botanicals, bioagents and chemical against ChLCV. In total 9 treatments were considered viz neem leaf extract, mahogoni leaf extract, garlic extract, trichoderma +raw cow milk, lantana leaf extract, trichoderma, marigold leaf extract, malathion and raw cow milk, the performance of selected 9 treatments against ChLCV in case of disease incidence(%), percent disease index(PDI), growth attributing factor like plant height, number of branches, number, yield attributing factor such as number of first flowering, 50% flowering, yield (Kg/ha)/plant, number of white fly were discussed.

In case of disease incidence (%) of ChLCV, the highest percent disease incidence was recorded 14.45, 16.14 and 18.12 at 35, 55, 75 DAT respectively was recorded in T₉ (Marigold leaf extract). The lowest percent disease incidence was 4.95, 7.82 and 9.97 in T₄ (Malathion) at 35, 55, 75 DAT. In case of PDI, the highest PDI was 12.84, 14.63, 19.25 in T₉ with moderately susceptible reaction and the lowest was 3.77, 6.38 and 8.68 in T₅ (Malathion) 35, 55, 75 DAT respectively.

The symptoms are observed downward curling of leaves, light and vein clearing, puckering of leaflets, infested twig almost shows upward and downward curling leaves with aborted flower and aborted flower and misshapen fruit.

In the following study, there were significant yield reduction and disease reduction because of the application of different plant extract and natural extract. It was found that ChLCV infected plants were maximum when the yield loss was the highest.

The yield was the lowest in T₉ (Marigold leaf extract) and there was the highest number of white fly population while the highest was in T₅ (Malathion) and there was the lowest white fly population.

In view of the results the present study may be concluded as

- ❖ ChLCV produces leaf curl symptoms which causes vein clearing on young leaves and at early stage of infection upward curling of young and old leaves and stunting in most of the cases.

- ❖ The lowest disease incidence , the lowest percent disease index were recorded in T₄(Malathion) at 35,55,75 DAT .
- ❖ A positive correlation was found between yield loss and white fly association and there was also positive relationship with white fly association and disease incidence.
- ❖ There are significant variation was found in growth and yield attributing characters among different treatments in ChLCV where T₄(Malathion) and T₁(Neem leaf extract) recorded better performance aganist ChLCV .

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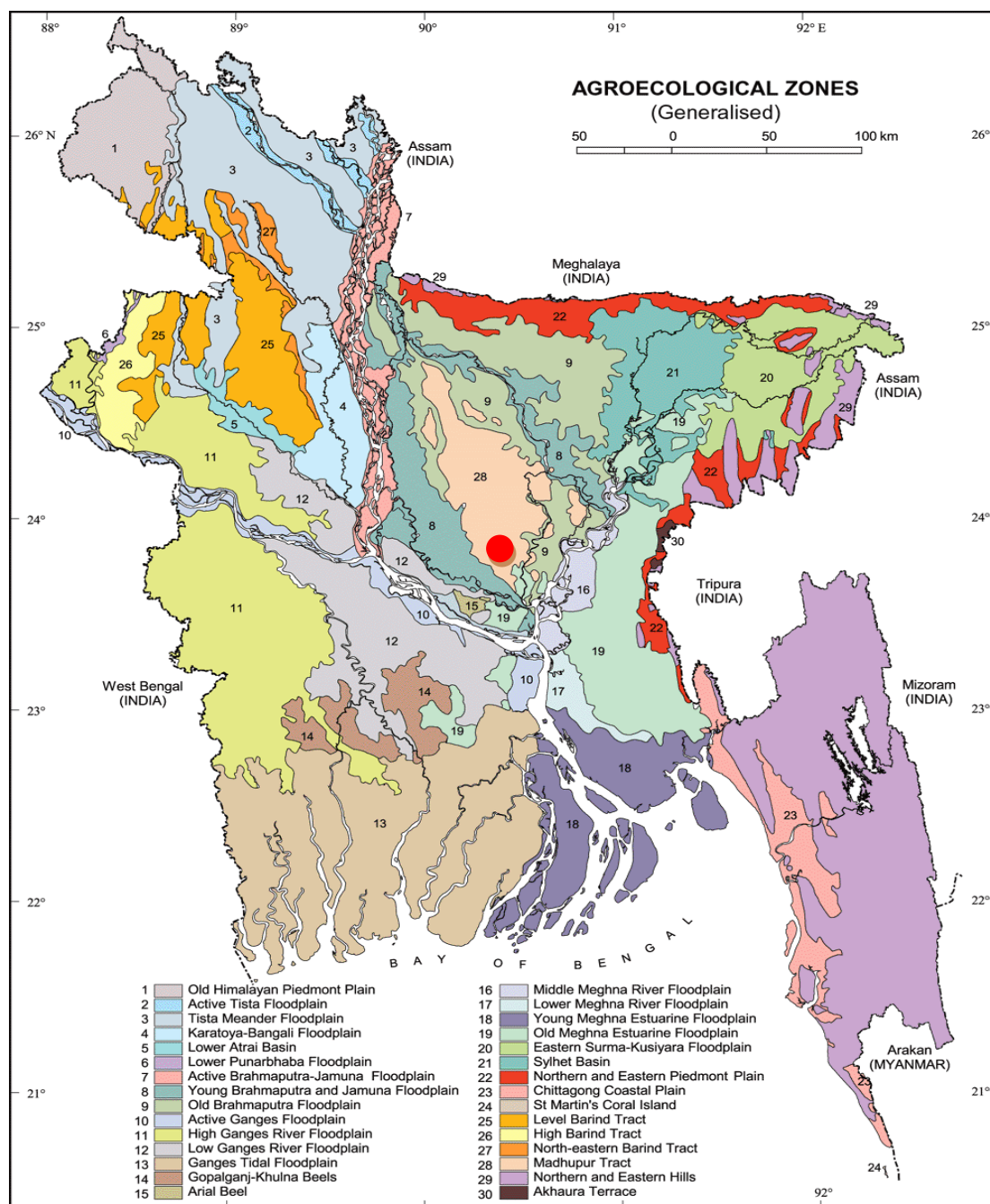
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APPENDICES

Appendix I. Experimental location on the map of Agro-ecological Zones of Bangladesh



Appendix II : The morphological and chemical characteristics of soil of experimental site as observed prior to experimentation

Morphological features	Characteristics
Location	Experimental field,SAU,Dhaka
AEZ	Modhupur tract(28)
General soil type	Shallow red brown terrace soil
Land type	Medium high
Soil series	Tejgaon
Topography	Fairly leveled
Food level	Above flood level
Drainage	Well drained
Texture	Loamy

Chemical composition

Constitutents	0-15 cm depth
p ^H	6.00-6.63
Total N(%)	0.07
Available P(µg/g)	18.49
Exchangeable K (µg/g)	0.07
Available S(µg/g)	20.82
Available Fe	229
Available Zn (µg/g)	4.48
Available Mg (µg/g)	0.825
Available Na(µg/g)	0.32
Available B (µg/g)	0.94
Organic matter(%)	0.83

Source : Soil Resources Development Institute (SRDI), Farmgate, Dhaka

Appendix III. Monthly mereological information during the period from December,2020 to April , 2020

Year	Month	Temperature (°C)		Relative humidity(%)	Total rainfall (mm)
		Maximum	Minimum		
2020	December	28.10	11.83	59.18	49
2021	January	26.00	10.53	70.45	00
	February	26.2	16.9	66	00
	March	32.89	20.2	57	24
	April	34.56	27.10	64.50	120

Source: Metrological Centre, Agargaon, Dhaka (Climate Division)