MANAGEMENT OF CERCOSPORA LEAF SPOT OF VEGETABLE COWPEA (*Vigna unguiculata* subsp. *Sesquipedalis* (L.) Verdcourt.).

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KERALA, INDIA

DECLARATION

I, hereby declare that this thesis entitled **"Management of Cercospora leaf spot of vegetable cowpea (*Vigna unguiculata* subsp. *Sesquipedalis* (L.) Verdcourt.)."** is a bonafide record of research work done by me during the course of research and the thesis has not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship or other similar title, of any other University or Society.

Place : Vellayani
Date : 3-11-2017

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CERTIFICATE

Certified that this thesis entitled "**Management of Cercospora leaf spot of vegetable cowpea (*Vigna unguiculata* subsp. *Sesquipedalis*(L.) Verdcourt.).** " is a record of research work done independently by Ms. Chinnu Ravi under my guidance and supervision and that it has not previously formed the basis for the award of any degree, diploma, fellowship or associateship to her.

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CHINNU RAVI

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LIST OF ABBREVIATIONS AND SYMBOLS USED

%	Per cent
µm	Micrometer
µl	Micro litre
µg	Microgram
@	At the rate of
°C	Degree Celsius
CD	Critical difference
cm	Centimeter
mm	Millimeter
et al.	And other co workers
Fig.	Figure
g	Gram
>	Greater than
h.	Hours
i.e.	that is
L	Litre
mL	Milli litre
DAS	Days After Sowing
Qt	Quintal
Kg	Kilo gram
min	Minutes
mg	Milli gram
sp. or spp.	Species (Singular and plural)
viz.	Namely
No.	Number
m^2	Meter square
aff	Affinity
F	Family

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INTRODUCTION

1. INTRODUCTION

Cowpea (*Vigna unguiculata* subsp *sesquipedalis* (L.) Verdcourt) is an important multi-season and multi-purpose legume vegetable that fits in to a variety of mixed farming systems in Kerala. It is a drought-tolerant, shade loving and warm-weather crop, well-adapted to Kerala conditions. The mature grains contain 23-25% protein, 50-67% starch, vitamin (folic acid) and contain essential micronutrients such as iron, calcium, and zinc (Abebe *et al.*, 2005).

The cultivation of cowpea is affected by a number of seed borne, soil borne and wind borne/foliar fungal diseases. Among the foliar diseases, affecting the crop, Cercospora leaf spots, powdery mildew and rust are of major concern (Ali *et al.*,2000). The Cercospora leaf spots appear as spots varying in colour and shape on the leaves and as lesions of stem and pods. In India, Cercospora leaf spot was first reported in Delhi, (Munjal *et al.*, 1960). Pant, (1989) reported severe incidence of the disease in cowpea grown at Varanasi. However, the disease remained a minor one until recent times. With the expansion of area under vegetable cowpea and cultivation of newer varieties coupled with intensive cultivation and changing climatic scenario would have accentuated the prevalence and severity of the disease .The disease now occupies a prime place among the various foliar diseases affecting the crop.

Two species of Cercospora have been reported to induce leaf spots in cowpea :*Cercospora canescens* and *Pseudocercospora cruenta* .In either case, air-borne spores produced on the underside of the leaf help in transmission. The fungus is carried over from one season to the next in left-over planting material. The fungus has a wide host range, attacking other legumes such as *Glycine max* (soybean), *Amaranthus tricolor*(grain amaranth), *Lablab purpureus* (hyacinth bean),*Mucuna pruriens* (buffalo bean) and the minor host include *Crotalaria juncea*(sunhemp), *Lycopersicon esculentum* (tomato) *Psophocarpus tetragonolobus,* (winged bean), *Vigna angularis* (adzuki bean), *Vigna mungo*

(black gram) and *Vigna radiata* (mung bean). These alternative hosts extend the reservoir of plants which can carry over infections to the next growing season.

Considering the increasing importance of this emerging pathogen in a remunerative crop like cowpea, there is urgent need to understand the symptomatology and etiology of the disease. A successful disease management strategy is also required to tackle the pathogen. In this context, the present study was taken up to study the details of the disease and pathogen and to develop an effective management strategy for Cercospora leaf spot. The objectives of the study was to understand the symptomatology of the disease and to find out the efficacy of fungicides, organic preparations and bio control agent on the management of Cercospora leaf spot under *in vitro* and *in vivo* conditions.

REVIEW OF LITERATURE

2. REVIEW OF LITERATURE

Cowpea *(Vigna unguiculata* subsp *sesquipedalis*) is a legume mainly grown in tropical and subtropical regions in the world for vegetable and grains and to a lesser extent as a fodder crop (Adegbite and Amusha, 2008). It has been recently observed that cowpea grown for vegetable purpose in Kerala is highly susceptible to leaf spot caused by *Cercospora* spp. leading to severe defoliation and consequent yield loss. Cercospora leaf spot (CLS) occurs in cowpea at any time of the year and can be severe in susceptible varieties. Pods and seeds also get infected (Allen *et al.*, 1998; Saxena *et al.*, 1998).

2.1. SYMPTOMATOLOGY AND ETIOLOGY OF CERCOSPORA

2.1.1. Symptomatology

Cercospora leaf spot caused by *Cercospora canescens* (Ellis and Martin) and *Pseudocercospora cruenta* (Sacc) formerly known *as Cercospora cruenta* (Sacc) have been observed in all the cowpea growing areas causing serious economic loss to the crop through sever defoliation (Emechebe and Shoyinka, 1985).

Symptoms of leaf spot caused by *C. canescens* on faba beans appeared predominantly as reddish brown to dark grey spots, sub-circular to angular, often concentrically zonate, with a broad, slightly raised, deep red margin (Yu, 1947).

Infection by *Pseudocercospora* sp appear as chlorotic spots on the upper leaf surface which gradually become necrotic with profuse black mat of conidia on the lower leaf surface. Symptoms caused by *C.canescens* appeared as rough, circular to irregular, 2-8 mm diameter, cherry red to dark red spots with a silvery grey centre and darker margin on the upper leaf surface with abundant conidia on the corresponding lower surface. (Emechebe and Shoyinka, 1985).

Cercospora canescens caused development of definite spots on leaves, which were brown at first, later turning grey or dirty grey with narrow reddish brown margin bearing fructification on both the surfaces of leaves of mung bean(*Vigna mungo* L.) (Munjal *et al.*, 1960).

As per the symptom description by Vasudeva (1963) the symptoms of leaf spot of faba beans caused by *C. canescens* initiated as brown spot which later became grey to dirty white, sub-circular to irregular of 5-10 mm width. The symptoms sometimes appeared on the stem, cotyledons and drying pod with black to greyish patches.

Vakli (1977) described the symptoms of Cercospora leaf spot of cowpea caused by *C. canescens* as round orange to light brown, silvery grey spots of 8-15 mm diameter. On the petiole, symptom appeared as uniform light brown spindle shaped lesion. Symptoms of *C. cruenta* appeared as brown coloured spots with circular to irregular dark margin which later became necrotic, grey coloured center with a brown margin. Cercospora leaf spot on faba beans mainly affects leaves, but also affect stems and pods (Lang *et al.*, 1993).

Patel *et al.* (2001) explained the symptoms of frog eye leaf spot of tobacco caused by *C. nicotianae*. The spots were brown with ash grey centers, gradually the centers of the spots may turn white and dry up. Typically the spots had a white center, surrounded by black margin resembling the eye of frog. Finally, several spots may coalesce towards the leaf tip and margin causing the leaf to dry up from the margin.

2.2.1. Etiology of the pathogen

The genus *Cercospora* was first proposed by Johann Baptist Georg Wolfgang Fresenius in 1863. The generic name was derived from the Greek word *kerkos* meaning tail and *spora* meaning spore, together called tail shaped spores. It was used for naming any Cercosporoid fungus, *ie.* a dematiaceous hyphomycete with filiform conidia (Pons and Sutton, 1985). *Cercospora* belongs to the subdivision of Ascomycete fungi, Class Dothidiomycetes, Order Capnodiale and Family Mycosphaerellaceae. The genus *Cercospora* is considered as one of the largest and most heterogeneous genera of hyphomycetes (Crous and Braun, 2003).

Verma and Patel (1969) reported that Cercospora leaf spot on cowpea appeared late in the season but progressed rapidly under conditions of moderate mean temperature (26°C) with 61-79 per cent of relative humidity and scanty rainfall (2.4 mm).

Mulder and Holliday (1975) described that the fruiting bodies of the pathogen are formed at 28°C. Pathogen survives mainly in plant debris as desiccation resistant pseudostromata, but can also survive as conidia in debris or in seeds and spread via rain-splash or wind to new leaves or plants. Sporulation is favoured by humid weather, warm temperature and dense plant population. High humidity of 95% or above was essential for conidial germination. Therefore, the disease is most severe during warm and moist weather. Conidia germinate over a wide range of temperature optimum being temperature 25-30 C ° (Kumar *et al.*, 2011).

The conidiophores damage the host tissue by rupturing the epidermal layer of the leaves or emerging out from the host tissue through stomatal openings (Meghvansi *et al.*, 2013).

2.3. MORPHOLOGY OF CERCOSPORA SP

The species concept and taxonomy of *Cercospora* sp are based upon morphological criteria, especially the dimensions and characteristics of the conidia such as length, width, base, and tip and characteristics of the conidiophores like length, diameter, geniculation, and fasciculation. Among this, the most important feature is the spore length and width (Chupp, 1954).

The *Cercospora* mycelium appeared to be very fine to coarse, fairly regular to very irregular, septate hyphae which for the most part ramify the host tissue.Chupp (1954) reported that the conidia of *C. canescens* are hyaline, obclavate, cylindrical, straight to curved, 5-18 septate with a conidial size ranging from 30-208 x 2.5-5 µm (length x width) with acute tip.

Chupp (1954) reported that the conidia of *C. cruenta* were hyaline to sub-hyaline, olivaceous brown, slightly curved to straight tip with acute ends, 5-9 septate. Gill and Singh (1962) reported that the conidia of *C. beticola* were hyaline, septate and measured from 4 x 54 to 165 µm.

Vasudeva (1963) in his compilation on "Indian Cercosporae" described the morphological characters of *C. nicotianae* as slender, slightly curved, thin walled, hyaline.3 to 6 septae measuring 40 to 75 x 3 to 5µm in size. He also described the conidial size of *C. canescens* as 100-120 x 5-6 µm, whereas the conidial size of *C.cruenta* ranged from 60-80 x 4 µm .Ellis (1976) described the conidial size of as *C. canescens* 50-150 x 3-3.5 µm while, conidial size of *C cruenta* ranged from 40-150 x 3-4.5 µm

Hennebert and Sutton (1994) observed that the most important features among the conidial morphology was the presence of thickened hilum, which indicates the point of attachment to the conidiophores. The conidial shape of *Cercospora* appeared straight to curved, filiform, obclavate to cylindrical, with

round to distinctly truncate base and sub acute tip. Colour of conidia was hyaline to pale olivaceous (Singh, *et al* .,1998; Crous and Braun, 2003).

2.3. IMPORTANCE AND YIELD LOSS

Cercospora leaf spot (CLS) occurs in cowpea at any time of the year and can be severe in susceptible varieties. Pods and seeds also get infected (Allen *et* al., 1998; Saxena *et al.*, 1998).Although the disease occurs mainly on cowpeas and other grain legumes, several other major and minor hosts have been identified. The major hosts of the pathogen are G*lycine max* (soybean), sesame, Sugar beet, Safflower, *Amaranthus tricolor* (grain amaranth), *Lablab purpureus* (hyacinth bean), *Mucuna pruriens* (buffalo bean) and the minor hosts include *Crotalaria juncea* (sun hemp), *Lycopersicon esculentum* (tomato) *Psophocarpus tetragonolobus,* (Winged bean) *Vigna angularis* (Adzuki bean), *Vigna mungo* (black gram) and *Vigna radiata* (Mung bean).

C. cruenta was first reported in United States on the species of *Phaseolus*, *Dolichos* and in other legumes (Butler, 1973). In India, the leaf spot was reported first in Delhi (Munjal *et al.*,1960) and is found to occur in all parts of the humid tropical areas of India, Bangladesh, Indonesia, Malaysia, Philippines, Taiwan as well as in Thailand (Pandey *et al.,* 2009).

Cercospora leaf spot is an important constraint to cowpea production, in the humid tropics (Schneider *et al.*, 1976). Yield loss attributed to the disease in susceptible cowpea varieties varied between 36% and 42% (Schneider *et al.* 1976; Fery *et al.*, 1977). Cercospora leaf spot disease symptoms are not apparent until the time of flowering but can rapidly progress leading to premature defoliation. (Williams 1975). Severe infections can result in lesions developing on pods and stems (Mulder and Holliday, 1975; Williams, 1975; Vakili, 1977).

Chowdhury (1945) reported that reduction in yield due to this disease in cowpea was 5% in Assam. Bird and Maramorosch (1975) reported that *C. cruenta* and *C. canescens* caused severe leaf spotting and defoliation in cowpea at Ibadan. Williams (1977) reported Cercospora leaf spot caused considerable yield losses in cowpea fields in Nigeria. A study conducted by Schwartz and Pastor-Corrales (1989) in USA, reported that *M. cruenta* leaf spot of cowpea reduced the seed yield of the susceptible variety by 35.6%. In Varanasi, India, leaf spot caused by *M. cruenta* was found to cause serious disease in cowpea was reported by Pant (1989). Pandey and Pandey (2002) recorded that the yield loss caused by Cercospora leaf spot was 42 % while 25%, 10% and 3% loss was recorded in mature pod, seed number per pod and average weight per 100 seed respectively.

The yield loss of 22% due to tikka disease in ground nut caused by *C. arachidicola* was reported by Sundaram (1965). Puranik *et al.* (1973) also reported 51.2% loss in yield of groundnut due to cercospora leaf spot .Severe incidence of *Cercospora arachidicola* caused considerable yield loss of 45.76% in groundnut (Ghuge *et al.*, 1981).Yield loss (11-18%) due to severe incidence of Cercospora leaf spot of sesame was also reported by Kumar and Mishra (1992).

Irena *et al.* (2006) reported that leaf spot caused by *C. beticola* reported disease severity of 29.1 to 89.8% and cause considerable loss in sugar beet production. Chankaew (2011) reported yield loss of 50 % in mung bean due to Cercospora leaf spot caused by the fungus *C. canescens*. Yield losses due to Cercospora leaf spot in sugar beet was reported to be 40 per cent in Northeastern Colorado (Harveson, 2013).

2.4. MANAGEMENT OF CERCOSPORA LEAF SPOT OF COWPEA

A successful disease management strategy is the only solution by which the legumes have been protected from the yield-reducing effects of the pathogen making them to economic insignificance. The control of leaf spot disease has

become difficult as most of the cultivars are susceptible and no variety is absolutely resistant to the disease.

Singh *et al.* (2001) described that as there is low level of resistance to Cercospora leaf spot, the cultural practices and chemical control play an important role in its management. Cultural practices such as field sanitation, crop rotation, destruction of infected crop debris, and avoiding collateral hosts in the vicinity of the crop may help in reducing the incidence. Effective control of *Cercospora* usually involves the application of fungicides such as Mancozeb, carbendazim, copper oxychloride and benomyl.

2.4.1. Fungicides

Triazole fungicides are highly effective against different fungal diseases, especially many leaf- spotting fungi, powdery mildews and rusts (Brown *et al.*, 1986).

Dahmen *et al.* (1992) reported that difenoconazole and propiconazole inhibit the spore germination of *C. arachidichola.* This inhibition of spore germination was due to the inhibition of cytochrome P-45 enzyme affecting C-14 demethylation which was essential for the development of the fungal cell membrane ergosterol. Hemachandra (2007) evaluated the efficacy of fungicides against leaf spot of sugar beet caused by *C. beticola* under *in vitro* condition. He reported that the lowest spore germination was recorded with difenoconozole and saaf at 0.1% compared to control. Swamy (2010) evaluated the efficacy of triazole fungicides against *C. capsici* infecting chili under *in vitro* condition. Among the fungicides, complete inhibition of mycelial growth was observed in propiconazole followed by hexaconazole (98.7%).

West (1983) reported that propiconazole gave good control of Cercospora leaf spot caused by *Mycosphaerella musicola.* Das and Mohanty (1986) reported that the plant treated with carbendazim (0.05%) recorded the lowest disease

intensity of 24.3% and the highest disease control of 65.00% against Cercospora leaf spot in green gram and the grain yield of green gram was 8.9 and 8.0 q/ha in the two consecutive years.

Vaishnav *et al.* (1992) revealed that two sprays of carbendazim (0.025%) or mancozeb (0.2 %) gave effective control the *Cercospora* leaf spot of sesame. Reddy *et al.* (1992) reported that two sprays of carbendazim @ 0.04 % or 0.025 % were the most promising and economical in getting effective control of frog eye spot leading to higher yield in tobacco. The minimum mean disease intensity (36.4 %) and maximum disease control (61.01 %) was recorded with the foliar application of 0.0125 per cent difenoconazole followed by propiconazole, hexaconazole, carbendazim and mancozeb in cowpea aganist Cercospora leaf spot (Kapadiya and Dhruji, 1999).

Thammaih (2003) reported propiconazole and difenoconazole were found effective against Sigatoka leaf spot caused by *C. musae*. Foliar application of propiconazole (0.15%) and carbendazim reduce the intensity of *C. canescens* in mung bean under greenhouse (Singh *et al.,* 2012). Sunkad and Mishra (2005) reported that the triazole fungicides such as propiconazole, hexaconazole and difenoconazole at 0.1 per cent recorded the lowest per cent disease index of 71.59 % 66.93%, 65. 21% respectively against cercospora leaf spot in groundnut.

Two foliar applications of carbendazim (0.1%) was effective against Cercospora leaf spot of mungbean (Tiwari and Kotasthane, 1984; Kaur *et al.*, 2004; Saxena and Tripathi, 2006). Spraying with either hexaconazole (0.1%) or propiconazole (0.1%) or carbendazim (0.05%) were found to be better in the management of the frog-eye leaf spot of tobacco in Karnataka (Hundekar *et al.*, 2005; Jahagirdar and Hundekar, 2010). Swamy (2010) recorded that the fungicides propiconazole and carbendazim at 0.1 per cent concentration sprayed for two times at 10 days interval were most effective in reducing the disease with per cent disease index of 11.67 and 16.48 respectively against Cercospora leaf spot of chilli.

Hemachandra *et al.* (2011) observed that the fungicides such as copper oxychloride, copper hydroxide, mancozeb and metiram at 0.25%, chlorothalonil Tridemorph, difenoconazole at (0.2%) and a combination product Carbendazim + Mancozeb at 0.1 percent recorded inhibition aganist *C.beticola* over control. Palakshappa *et al.* (2011) conducted an experiment during *Kharif* 2009 at Main Agricultural Research Station, University of Agricultural Sciences, Dharwad and revealed that the fungicides *viz.,* Quintal and Carbendazim 0.1% recorded lowest Percent disease incidence of 44.41 and 48.50 and recorded highest yield of 470 kg / ha and 352 kg / ha respectively.

Singh *et al.* (2013) noticed that carbendazim(0.1%) sprayed at two times reported the minimum disease intensity (14.45%) and maximum (76.50%) disease reduction over the control in mung bean under green house condition.

Veena *et al.* (2014) reported that foliar application of difenoconazole (0.1%) gave superior pod number and fruit yield when compared to untreated control in green gram. Bhat *et al.* (2015) reported that the foliar spray of carbendazim (0.05%) and difenoconazole(0.02 %) significantly reduced the pod infection (77 %) leaf spot intensity (68 %) of *C. canescens* of green gram and gave an attractive seed yield of 8.62-8.95 q/ha at an acceptable B:C ratio (6.38:1)

2.4.2. **Organic formulations**

Botanicals have effective antifungal activity against many plant pathogens. They act as an ideal source of low cost and eco-friendly management of plant diseases. The inhibitory effects of the plant products on pathogen are attributable due to the presence of some antifungal ingredients (Singh and Dwivedi, 1987). Khan and Rishi Kumar (1990) studied antifungal activity of leaf extract of neem (*Azadirachta indica*). Botanicals, particularly neem (*A. indica*) and vegetable oils have shown good fungicidal potential against many foliar pathogens of different crops as eco friendly fungicide (Ranga Rao *et al.*, 2007).

Benagi (1995) evaluated 22 plant extracts against the conidial germination of C. *personata*. Out of which leaf extracts of *A. indica* and neem seed kernel extract (NSKE) exhibited maximum inhibition of conidial germination at 2.5 per cent. Leaf and kernel extract of *A. indica* at 2.5 % was found inhibitory to conidial germination of *Phaeoisariopsis personata*. Khadar (1999) evaluated 22 plant extracts against *C. arachidicola*. Among the extracts, maximum per cent inhibition of conidial germination was recorded in neem leaf and kernel extract. Swamy (2010) evaluated nine botanicals *in vitro* and observed significantly higher inhibition of mycelial growth in neem seed kernel extract (87.87%).

Studies conducted by Thammaiah and Shirol (2013) revealed that, three sprays of propiconazole 0.05%+ petroleum based mineral oil 1% effectively controlled(13.7 8 %) the Sigatoka leaf spot disease in Banana. The activity of this group of oils had been shown to delay symptom development and inhibit lesion growth by reducing spore germination, germ-tube growth and sporulation of the Sigatoka pathogen.

Neem oil, neem seed kernel extract and neem cake extract at 2.0 % found effective in controlling late leaf spot of groundnut caused by *Cercospora personatum* under field condition (Usman *et al.*,1991). Clarified hydrophobic neem oil (70 %) was found effective against Cercospora leaf spot of eggplant for controlling Cercospora leaf spot disease of eggplant (Srivastava and Nelson, 2012). Trivedi (2014) reported efficacy of neem oil aganist the Cercospora leaf spot in black gram caused by *C. canescens*. Under field condition, foliar spray of neem oil at 0.2% recorded the lowest percentage disease index of 29.38%. Kumar *et al*. (2017) reported that neem oil at 5% was found effective against Tikka leaf spot of groundnut caused by *Cercospora* sp.

2.4.3. Biological control by *Pseudomonas fluorescens*

Pseudomonas fluorescens have emerged as the largest and potential promising group of plant growth promoting rhizobacteria (PGPR) for the bio-control of plant diseases (Liu *et al.*, 1995). They acts through the production of hydrolytic enzymes, antibiotics, induction of plant host defense mechanisms, niche colonization, competition, and interference with pathogenicity factors (Punja and Utkhede, 2003).

Simonetti (2012) evaluated the *in vitro* efficacy of the biocontrol agents against leaf spot of soybean caused by *C. sojina* and found that *P.fluorescens* completely inhibited the mycelia growth and spore germination of the pathogen.

Raguchander *et al.* (2005) reported that the foliar spray of *P. fluorescens* at 2.0 per cent under field condition reduced the Cercospora leaf spot disease incidence by 94.80% in Urd bean. Pairashi *et al.* (2007) noticed the reduction in the incidence of leaf spot in tobacco caused by *C. nicotianae* by the application of *P. fluorescens* at the rate of 2 g/L.

Devappa and Thejakumar (2016) reported that the foliar application of Tilt 25% @ 0.05% + Eucalyptus @ 10% + *P. fluorescens* @ 5g/L effectively managed the Cercospora leaf spot disease with PDI range from 8.00 to 21.33 which was on par with treatment Tilt 25% @ 0.05% + *P. fluorescens*@ 5g/ L with a PDI range of 10.67 to 25.33 compared to control.

Poornima (2010) found that *P. fluorescens* at 0.2 per cent recorded maximum suppression of disease with least per cent disease index (4.16%) aganist leaf spot of palak caused by *C. beticola* followed by *T. harzianum* (0.2%). Foliar application of *P. fluorescens* (5%) gave a significant increase in the plant height, maximum number of leaves, pod yield and the minimum disease intensity of 20.1% in groundnut according to Kumar *et al.* (2017).

MATERIALS & METHODS

3. MATERIALS AND METHODS

The present study entitled "Management of Cercospora leaf spot of vegetable cowpea(*Vigna unguiculata* subsp *sesquipedalis*) was carried out in the Department of Plant Pathology, College of Agriculture, Vellayani and Coconut Research Station, Balaramapuram during the year 2015-2017. Details of materials used and the methods followed for the study are presented below.

3.1. SYMPTOMATOLOGY AND ETIOLOGY OF CERCOSPORA

3.1.1. Symptomatology

Symptomatology of the disease caused by *Cercospora* spp was studied by observing the naturally infected cowpea leaves, pods and stem. The diseased samples were collected from six different locations near to College of Agriculture, Vellayani. The symptoms caused by *Cercospora* spp in cowpea in different plant parts were assessed and recorded.

3.1.2. Etiological studies

3.1.2.1. *Conidial Characteristics of Pathogen*

For studying the conidial characteristics, scrapings were taken from the diseased leaf samples showing the symptoms of Cercospora leaf spot. Scrapings were placed in a clean slide containing a drop of lacto phenol cotton blue and observed under the compound microscope (40 X). Observations were recorded on conidial characteristics such as conidial colour, length, width and septation.

3.1.2.2. *Mycelial Characteristics of Pathogen*

Leaves showing the symptoms of *Cercospora* were collected from the field and then placed inside the moist chamber prepared by placing two moist filter papers in the inner surfaces of a petriplate. After few days, white mycelial

strands were visible on the upper surface of leaves. A bit of mycelium was taken out using needle and placed it on a clean slide containing a drop of lacto phenol cotton blue stain and observed under the microscope. Observations were recorded on mycelial characteristics such as the hyphal morphology, branching and septation.

3.1.2.3. *Identification of the Pathogen*

Cowpea leaves showing well developed symptoms of Cercospora leaf spot were collected from College of Agriculture, Vellayani. The collected leaves were pressed in between news papers for 10 days by changing the paper at every alternate days untill the leaves had completely dried out. Later, the dried leaves were properly packed in paper bags and was send to National Fungal Culture Collection of India, (N.F.C.C.I) Pune for identification. The identity of the pathogen was confirmed by the morphological characterization undertaken at N.F.C.C.I, Pune.

3.2. EVALUATION OF THE EFFICACY OF FUNGICIDES, ORGANIC FORMULATIONS AND BIO CONTROL AGENT ON INHIBITION OF *Cercospora* sp

An experiment was conducted under *in vitro* conditions in Completely Randomized Design (CRD) with three replications to evaluate the efficacy fungicides, organic formulations and bio-control agent on inhibition of *Cercospora* sp by spore germination assay. The concentration of fungicides, organic formulations and bio-control agent used in the study are presented below

Fungicides	Trade name	Concentrations
Tebuconazole	**Folicur**	**0.05 , 0.1%**
Propiconazole	**Tilt**	**0.05, 0.1%**

Azoxystrobin	**Amistar**	**0.05 , 0.1%**
Hexaconazole	**Mesamaster**	**0.05 , 0.1%**
Difenoconazole	**Score**	**0.05 , 0.1%**
Carbendazim	**Megastin**	**0.05 , 0.1%**
Copper hydroxide	**Kocide**	**0.15% ,0.2%**
Mancozeb	**Indofil M-45**	**0.3% ,0.4%**
ORGANIC FORMULATIONS		
Panchagavya	**-**	**5.0%**
Fish aminoacid	**-**	**5.0%**
Neem oil	**-**	**0.5%**
Mineral oil	**-**	**0.1%**
BIOCONTROL AGENT		
Pseudomonas fluorescens	**KAU talc based formulation**	**2.0%**

3.2.1. Preparation of Conidial Suspension

Cowpea leaves showing characteristics symptoms of Cercospora leaf spot were collected and kept in humid overnight for inducing sporulation . One ml of sterile water was poured on a severely infected leaf and was washed out to get a rich suspension of conidia. The conidial suspension thus obtained was mixed with sterile water taken in a petriplate and average count was taken using haemocytometer. Later the conidial suspension of the fungus was adjusted to a desired concentration of 50 conidia/ mL and was mixed separately with the different concentrations of treatments (Imtaij *et al*., 2005)

3.2.2. Preparation of Panchagavya

Panchagavya was prepared by following the procedure described in organic POP, KAU (2009). Initially, fresh cow dung (7 kg) and cow ghee (1 kg) were mixed thoroughly in a clean container and was kept aside for three days. The mixture was stirred thoroughly twice a day during morning and evening hours. After three days, cow urine (10 L) and water (10 L) were added to this mixture and was kept aside for another 15 days, with regular mixing twice a day. After 15 days, cow milk (3 L), cow curd (2L), tender coconut water (3L), jaggery (3 kg) and well ripened poovan banana (12 Nos.) were added to the above mixture. The mouth of the container was properly covered and the container was kept under shade with regular stirring. Panchagavya was ready for use after 30 days and was used at the rate of five per cent concentration.

3. 2.3. Preparation of Fish amino acid

Fish amino acid was prepared by following the steps described by Weinert *et al* .(2014) by mixing one kg of Sardine fish (*Sardina pilchardus*) with one kg of jaggery in a plastic can and kept it under shade condition. The mouth of the can was covered with paper and tied with string and kept undisturbed for 25 days. After 25 days, the content was filtered through muslin cloth and stored in the same can. The filtered content was used for further studies.

3.2.4. *In vitro* evaluation of fungicides, organic formulations and bio control agent against *Cercospora* sp by conidial germination assay

The fungicides, organic formulations and biocontrol agent used in the study were evaluated *in vitro* against *Cercospora* sp by conidial germination assay using Hanging Drop Technique (Usha and Rekha, 2014). Conidial suspension of fungus containing 50 conidia per microscopic field was prepared. One drop (100 ul) of spore suspension was placed in a cavity slide containing a drop (100 ul) of different concentrations of the treatment and a cover slip was placed over the slide. The slides were then placed in moist chamber and were incubated at room temperature for 24 h. Conidia mixed in sterile water served as the control. Three replications were made for each treatment. The slides were observed under a light microscope (40X) for recording the germinated conidia. The conidial germination was calculated using the formula developed by Kiraly *et al.* (1974)

$$\% \text{ conidial germination} = \frac{\text{Number of conidia germinated}}{\text{Total number of conidia examined}} \times 100$$

3.3 MANAGEMENT OF CERCOSPORA LEAF SPOT OF VEGETABLE COWPEA IN POT CULTURE STUDIES

Based on the results of *in vitro* evaluation a pot culture experiment was laid out in C.R.D. at Coconut Research Station (CRS), Balaramapuram to find the efficacy of the fungicides, organic formulations and biocontrol agent against *Cercospora* sp. The details of experiment were as follows; T1 : Propiconazole (0.1%), T2 : Azoxystrobin (0.1%), T3 : Hexaconazole (0.1%), T4 : Difenoconazole (0.1%), T5 : Carbendazim (0.1%),T6 : Neem oil (0.5%,) T7 : Mineral oil (0.1%) (Petroleum based), T8 : *P. fluorescens* (2.0 %) (KAU talc formulation), T9 : Inoculated control

Cowpea seeds of the variety Vellayani Jyothika were sown in grow bags of size 40 x 24 x 24 cm^3 which were filled with potting mixture (sand, soil and cow dung) in the ratio 1:1:1. Each grow bag contained two plants and three replications were maintained for each treatment

3.3.1. Application of inoculum

Artificial dusting of infected leaf samples collected from the field and mulching the base of the cowpea plants with 10g of infected leaf samples were followed .

3.3.2. Application of Fungicides

Fungicides application was done thrice at 35, 50 and 65 DAS (Days after sowing) as foliar spray.

3.3.3. Application of Organic Formulations

Organic formulations like mineral oil (0.1 per cent) and neem oil (0.5 per cent) was prepared and applied as foliar spray thrice at 35, 50 and 65 DAS as foliar spray

3.3.4. Application of *Pseudomonas fluorescens*

KAU talc based formulation of *P. fluorescens* (2.0 per cent) was prepared and applied as foliar spray thrice at 35, 50, 65 DAS as foliar spray.

3.3.5. Observations

The following observations were recorded on 45, 60 and 75 DAS.

3.3.5.1. *Number of Days taken for the Appearance of Cercospora Leaf Spot*

The plants were observed for the first appearance of the disease following the artificial inoculation of the pathogen.

3.3.5.2. *Disease Incidence (%)*

The incidence of disease was calculated according to the formula of Agrios (2005)

$$\text{Disease incidence} : \frac{\text{Number of infected leaves}}{\text{Total number of leaves}} \times 100$$

3.3.5.3 *Disease Index*

Based on the extent of damage caused by Cercospora leaf spot, disease severity was assessed at 10 days interval using a 0-5 scale (Plate 1) developed by (Oladiran, 1983) as follows :

Grade	Description
0	Healthy
1	1-10 per cent of leaf area infected
2	11-25 per cent of leaf area infected
3	26-50 per cent of leaf area infected
4	51-75 per cent of leaf area infected
5	more than 75 per cent of leaf area infected

Plate 1.Score chart of Cercospora leaf spot of cowpea

Based on the scores assigned to each diseased plant/leaf, severity (disease index) was worked out using the formula described by Wheeler (1969)

$$\text{Disease severity} : \frac{\text{Sum of grades of each leaf} \times 100}{\text{Total no of leaves assessed} \times \text{Maximum grades used}}$$

3.3.4.4. *Yield characters*

Yield of the cowpea (kg/pot) was obtained by picking the pods at an interval of two days. Number of pods and the weight of the pods obtained from each pot were recorded and pooled.

3.4. FIELD EVALUATION OF SELECTED FUNGICIDES, ORGANIC FORMULATIONS AND *P. fluorescens* FOR SUPPRESSION OF CERCOSPORA LEAF SPOT

Based on the results of pot experiment, two effective fungicides, two organic formulations and *P. fluorescens* were selected to evaluate under field conditions along with the chemical control check carbendazim (0.1%). The details of the experiment are as follows ; Design-RBD, Treatments-7, Replication- 3, Variety – Vellayani Jyothika , Plot size- 5x4 m^2, Season –Oct to Dec, Location- Coconut Research Station, Balaramapuram. The crop was raised during the period from February 2017 to June 2017.The treatment details for the experiment were as follows T1 : Propiconazole (0.1%), T2 : Difenoconazole (0.1%) T3 : Neem oil (0.5%), T4 : Mineral oil(0.1%) (Petroleum based), T5 : *P. fluorescens* (2.0 %) (KAU talc formulation), T6: Chemical control check (Carbendazim 0.1%), T7: Control.

The plot was manually leveled and arranged in Randomized Block Design (RBD) with three replications. Seeds were dibbled at a row spacing of 50 cm and plant to plant spacing was 30 cm. Fertilizer application and cultural operations

have been done as per the Package of Practices Recommendations of crops, KAU (2009).

Treatments application was done thrice at an interval of 15 days starting from 35, 50, 65 days after sowing following the natural unprotected field conditions. Observations on number of days, Disease incidence, Disease index, Pod yield (kg /plot) were recorded as described in 3.3.5.

3.4.1. Incidence of other diseases and pest

The infestation by the other pest and disease was observed periodically during 45, 60 and 75 DAS and recorded.

3.5. BENEFIT: COST RATIO

The weight of pods harvested was recorded and expressed as kg/plant and converted to kg/ha. The parameters *viz.*, cost of cultivation, labour cost and cost of plant protection were calculated to compute the benefit cost ratio. The benefit cost ratio was obtained by monetary benefits by additional cost incurred on each treatment.

3.6. STATISTICAL ANALYSIS

Data obtained from the experiment were subjected to statistical analysis applying ANOVA technique was tested by F test. In the cases where the effects were found to be significant, critical difference values were calculated for each observation using table t values at 5 per cent level of significance. Then, the significance of treatments were compared with critical difference values.

RESULTS

4. RESULTS

The experiments were conducted under laboratory, pot and field conditions during the 2015-2017 at Department of Plant Pathology, College of Agriculture, Vellayani and Coconut Research Station, Balaramapuram. The results obtained from the experiment are summarized below.

4.1. SYMPTOMATOLOGY AND ETIOLOGY OF CERCOSPORA LEAF SPOT

4.1.1. Symptomatology

Symptomatology of the Cercospora leaf spot was studied by observing the infected samples from the field near College of Agriculture,Vellayani. The symptoms were observed on the leaves, pods and stem. Infection by *Pseudoercospora cruenta* on leaves, appeared intially as small light greenish yellow diffused spots on the adaxial surface of the leaf. Later these spots turn reddish brown circular to angular, 2-10 mm diameter with a greenish yellow halo. As the spot enlarged, the leaves showed yellowing and black mass of conidia were seen on the upper and lower surface of leaves. Later they became completely chlorotic and the spots were bordered by water soaked greenish halo. At the final stage, the leaves became necrotic, dried up and hung down. (Plate 2)

On the stem, symptom appeared as black coloured lesions with mass of conidia (Plate 3). In pods, irregular black spots appeared with powdery mass of fungal growth on and inside the fruits, damaging them completely. (Plate 4)

Symptoms of *Cercospora canescens* on leaves appeared as rough, diffused and irregular, dark red spots with a silvery grey centre and a darker margin. Conidia were produced on the corresponding lower surface of the leaves. (Plate 5)

Stage 1

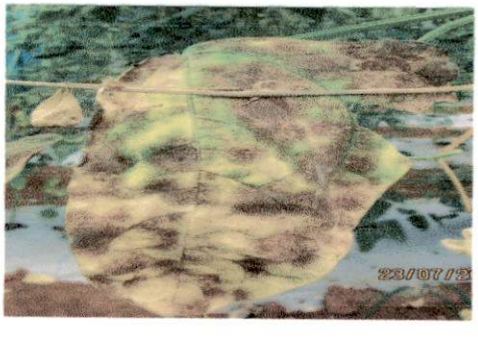
Stage 2

Stage 3

Stage 4

Stage 5

Stage 6



Stage 7

Stage 8

Plate 2. Different stages of symptoms of *Pseudocercospora cruenta* on leaves

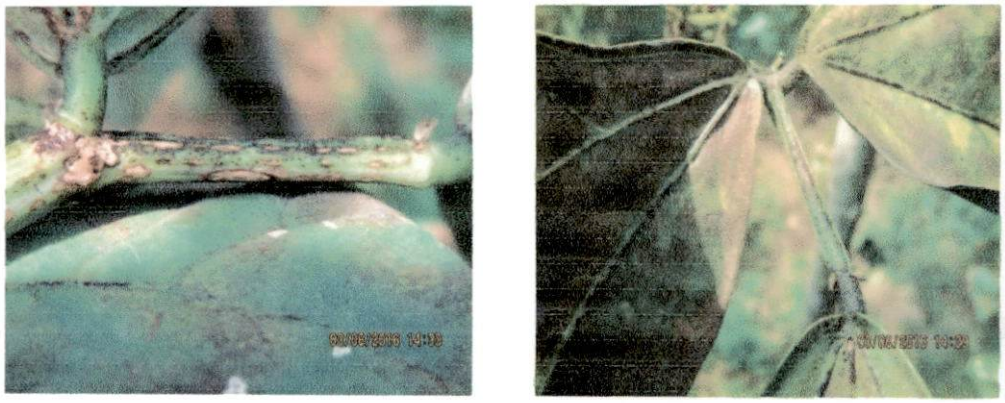


Plate 3. ***Pseudocercospora cruenta*** **symptom on stem**

Plate 4 . ***Pseudoercospora cruenta*** **symptom on pods**

Plate 5. ***Cercospora canescens*** **symptom on leaves**

4.1.2. Etiology

4.1.2.1. Conidial characters

Conidial characters such as conidial length, width, colour and septation were recorded by microscopic studies and the data was depicted in the Table 1, Plate 6.

Conidia produced from the diseased leaf samples were hyaline, obclavate, straight to curved tip and septate with conidial length ranging from 24.7 to 57.6 µm and width of 1.1 to 2.7µm. The average size of the conidia was maximum (57.6 x 2.7 µm) for the diseased leaf samples collected from Balaramapuram followed by Kakkamoola (55. 2 x 2.1 µm) . The minimum average size was recorded with leaf samples collected from Poonkulam (24.64 x 1.74µm). Maximum Conidial septation ranged from 10 to 17 in number.

4.1.2.2. Mycelial characters

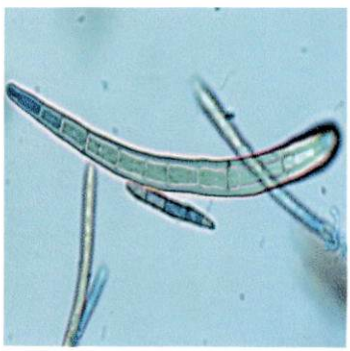
Mycelia of the pathogen was very fine to coarse, fairly regular to very irregular, well developed, branched, intercellular with septate hyphae. (Plate 7)

4.1.2.3. Identification of the pathogen

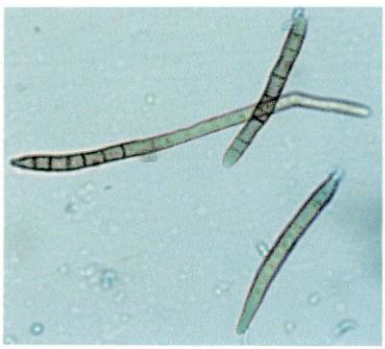
Based on the conidial and mycelial characters, the pathogen was tentatively identified as *Cercospora* sp. The identity of the pathogen was further confirmed based on the morphological characteristics and the herbarium specimen send to National Fungal Culture Collection of India, (N.F.C.C.I), Pune as *Pseudocercospora* sp. aff. *Pseudocercospora vignigena* (F: Mycosphaerellaceae) Accession No: 499472.

Table 1. Microscopic observations of different samples of *Cercospora* sp obtained from surveyed locations

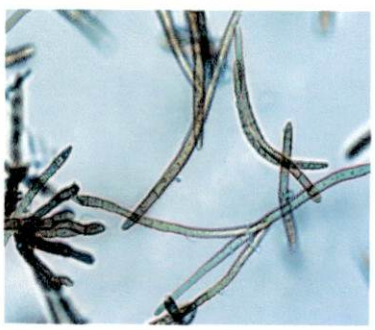
	CONIDIAL CHARACTERS			
Locations	**Colour of the conidia**	**Shape of conidia**	**Mean conidial dimension (length *breadth) (µm)**	**Conidial septation**
LOCATION 1 Kakkamoola.	**Hyaline**	**Obclavate , straight to curved tip**	**55.2 x 2.1**	**12-14**
LOCATION 2 Balaramapuram	**Hyaline**	**Obclavate , straight to curved tip**	**57.6 x 2.7**	**15-17**
LOCATION 3 Vellayani	**Hyaline**	**Obclavate, straight to curved tip.**	**39.4 x 2.1**	**11-15**
LOCATION 4 Kalliyoor	**Hyaline**	**Obclavate, straight to curved tip.**	**47.24 x 1.06**	**15-16**
LOCATION 5 Poonkulam	**Hyaline**	**Obclavate, straight to curved tip.**	**24.64 x 1.74**	**10-14**
LOCATION 6 Manakadu	**Hyaline**	**Obclavate, straight to curved tip.**	**46.57 x 2.02**	**12-16**



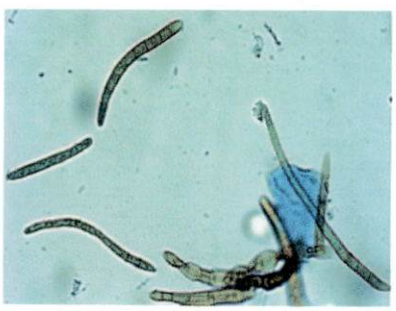
Kakkamoola



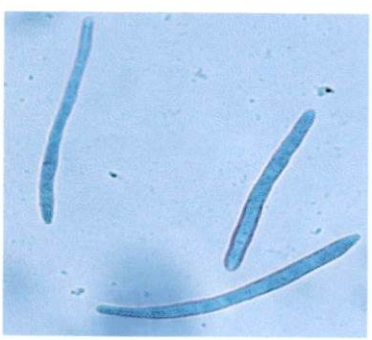
Balaramapuram



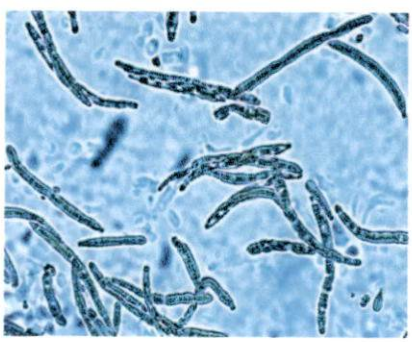
Vellayani



Kalliyoor



Poonkulam



Manakadu

Plate 6. Conidia of different samples collected

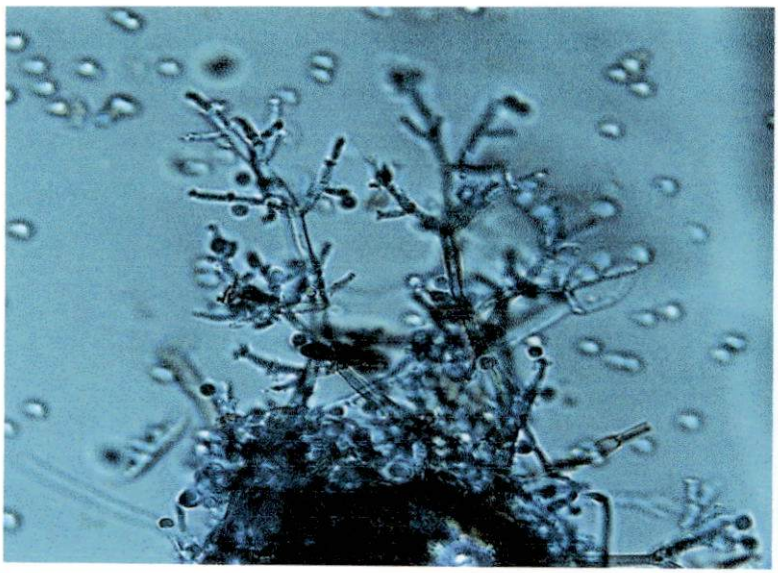


Plate 7 .Mycelia of pathogen

4.2 Evaluation of the fungicides, organic formulations and *Pseudomonas fluorescens* on inhibition of *Cercospora* sp.

The results of efficacy of fungicides, organic formulations and *P. fluorescens* on the conidial germination of *Cercospora* sp are furnished in Table 2, (Plate 8).

The results revealed that all the fungicides tested *in vitro* aganist *Cercospora* sp significantly inhibited the conidial germination of pathogen at two concentrations over the control. Of the fungicides tested, difenoconazole (0.1%) recorded the lowest percentage (53.87 %) of conidial germination with 42.96% reduction over the control and differed significantly from all other treatments. This was followed by propiconazole (0.1%) with 59.63% of conidial germination which was on par with hexaconazole (0.1%) with 65.06 %conidial germination. Conidial germination was recorded highest (83.66%) in copper hydroxide (0.1%) with 11.41% reduction over the control.

In the case of organic formulations, mineral oil (0.1%) and neem oil (0.5%) recorded 73.77% conidial germination with 21.88 % inhibition of conidial germination over control and it was statistically on par with each other. Maximum conidial germination of 85.10 % was observed with panchagavya (5%) and followed by fish amino acid (82.77%) and were on par with each other.

The recommended dosage of KAU talc based formulation of *P. fluorescens* recorded only 11.88 % reduction of conidial germination over control and it was on par with fish amino acid which recorded 12.35 %.

4.3. Evaluation of fungicides, organic formulations and *P. fluorescens* on Cercospora leaf spot management under pot culture conditions

A pot culture study was conducted at Coconut Research Station, Balaramapuram to evaluate the five best fungicides (difenoconazole (0.1%), propiconazole (0.1%), azoxystrobin (0.1%), carbendazim (0.1%), hexaconazole

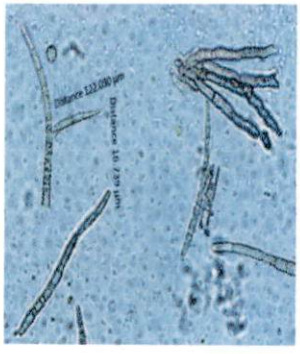
Table 2. Effect of fungicides, organic formulations and *Pseudomonas fluorescens* on inhibition of conidial germination of *Cercospora* sp

Treatment No	Treatments	Conidia germination (%)	Inhibition of conidia germination (%)
	FUNGICIDES		
T1	Tebuconazole (0.1%)	78.44 (62.54) ^{bcd}	16.94
T2	Propiconazole (0.1%	59.63 (50.58) ^{g}	36.85
T3	Azoxystrobin (0.1%)	77.10 (61.45) ^{cde}	18.36
T4	Hexaconazole (0.1%)	65.06 (53.79) ^{fg}	31.10
T5	Difenoconazole (0.1%)	53.86 (47.22) ^{h}	42.96
T6	Carbendazim (0.1%)	70.00 (56.79) ^{ef}	25.87
T7	Copper hydroxide (0.2%)	83.66 (66.25) ^{bc}	11.41
T8	Mancozeb (0.4%)	79.21 (62.96) ^{bcd}	16.12
	ORGANIC FORMULATIONS		
T9	Panchagavya (5%)	85.106 (67.38) ^{b}	9.88

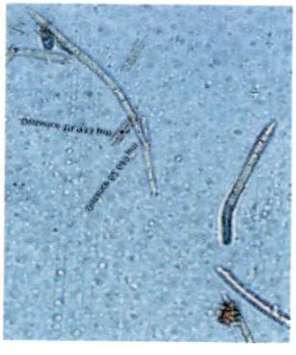
T10	**Neem oil (0.5%)**	**73.773 (59.25) ^{de}**	**21.88**
T11	**Fish aminoacid (5%)**	**82.776 (65.56) ^{bc}**	**12.35**
T12	**Mineral oil (0.1%)**	**73.773 (51.66) ^{fgh}**	**21.88**
	BIO CONTROL AGENT		
T13	***P. fluorescens* (2%)**	**83.220 (65.88) ^{bc}**	**11.88**
	Control	**94.440 (76.37) ^{a}**	
	CD(0.05)	**5.134**	

Mean of three replications, values in the parenthesis are arc sin transformed.
Treatments with same alphabets in the superscript do not differ significantly

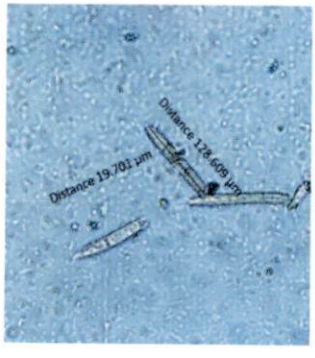
Difenoconazole (0.1 %)



Propiconazole (0.1 %)



Mineral oil (0.1%)



Neem oil (0.5%)

Pseudomonas fluorescens (2.0%)

Control

Plate 8. Conidial germination assay under *in vitro* condition

Plate 9. General view of pot culture experiment

(0.1%), two best organic formulations (mineral oil (0.1%), neem oil (0.5%) and *P. fluorescens* (2%) on Cercospora leaf spot management selected from *in vitro* evaluation (Plate 9). The following observations were recorded

4.3.1. Number of days taken for Cercospora leaf spot appearance

On artificial inoculation with the infected leaf samples Cercospora leaf spot appeared after 17 days in all the plants.

4.3.2. Disease incidence

The statistical data on disease incidence (Table 3) estimated at 45 DAS revealed that disease incidence in plants ranged from 17.16 to 33.47 per cent. Of the fungicides tested difenoconazole (0.1%) recorded the lowest (17.16 %) disease incidence with 51.74 % disease suppression over the control. This was on par with mineral oil (0.1%) with 23.17% disease incidence and 52.05% disease suppression over control. This was followed by propiconazole (0.1%) with 23.32% disease incidence and hexaconazole (0.1%) with 24.43 % of disease incidence. Among the treatments, highest (33.47%) disease incidence was observed in azoxystrobin (0.1%) with only 30.74 % disease suppression over the control.

Disease incidence estimated at 60 DAS revealed that the least incidence was recorded with difenoconazole (0.1%) and propiconazole (0.1%) sprayed plants with 22.18 % and 22.80% disease incidence respectively and they were found to be statistically on par. This was followed by hexaconazole (0.1%), mineral oil (0.1%) and carbendazim (0.1%) with 24.84%, 25.85%, 27.33% disease incidence respectively and were statistically on par with each other. Disease incidence was recorded highest (38.04%) in azoxystrobin (0.1%) and was on par with neem oil (0.1%) and *P. fluorescens* with 32.22 % and 33.96 % disease incidence respectively.

Table 3. Effect of fungicides, organic formulations and *P. fluorescens* in leaf spot disease incidence of vegetable cowpea in pot culture studies

		Mean disease incidence (%)			Percentage reduction of disease incidence over control		
Trt no	Treatments	45 DAS	60 DAS	75DAS	45DAS	60DAS	75 DAS
T1	Propiconazole (0.1%)	23.32 (28.69) ^{c}	22.80 (28.40)^{e}	26.83 (31.03) ^{de}	51.74	52.82	49.05
T2	Azoxystrobin (0.1%)	33.47 (35.34) ^{b}	38.04 (38.02) ^{b}	40.833 (39.70) ^{bc}	30.74	21.29	22.45
T3	Hexaconazole (0.1%)	24.43 (29.55) ^{e}	24.84 (29.89)^{de}	27.33 (31.47) ^{de}	49.45	49.96	48.10
T4	Difenoconazole (0.1%)	17.16 (24.45) ^{d}	22.18 (27.84)^{e}	23.80 (29.10) ^{e}	64.49	54.10	54.80
T5	Carbendazim (0. 1%)	25.45 (30.27) ^{e}	27.33 (31.47)^{cde}	31.09 (33.88) ^{de}	47.34	43.45	40.96
T6	Neem oil (0.5%)	30.19 (33.32) ^{bc}	32.22 (34.57) ^{bcd}	32.70 (34.87) ^{cd}	37.53	33.33	37.90
T7	Mineral oil (0.1%)	23.17 (28.76) ^{cd}	25.85 (30.541)^{cde}	30.046 (33.19) ^{de}	52.05	46.51	42.95
T8	*Pseudomonas fluorescens* (2%)	28.07 (31.84)) ^{bc}	33.96 (35.60) ^{bc}	41.64 (40.14) ^{b}	41.92	29.73	20.92
T9	Control	48.33 (44.04))^{a}	48.33 (44.046)^{a}	52.66 (46.53) ^{a}			
	CD(0.05)	4.758	5.61	5.227			

mean of three replications, values in the parenthesis are arc sin transformed.
Treatments with same alphabets in the superscript do not differ significantly

30 52

At 75 DAS, among the fungicides the lowest (23.80%) disease incidence was recorded with difenoconazole (0.1%) with 54.80% of disease suppression over the control which was on par with propiconazole 0.1%(26.83%), hexaconazole 0.1%,(27.33%) and carbendazim (0.1%)(31.09%).The highest (40.83%) disease incidence was recorded by azoxystrobin with 22.45% disease suppression over control and differed significantly from other fungicides.

Mineral oil (0.1 %) showed the lowest (30.04%) disease incidence among organic formulations with 42.95% disease suppression over the control and was statistically on par with neem oil (0.5%) with 37.9 % disease suppression over control.

Among all the treatments, *P. fluorescens* recorded the highest (41.64%) disease incidence with only 20.92% disease suppression over the control and was on par with azoxystrobin(0.1%). However, the untreated control plants recorded the highest (52.66%) disease incidence and differed significantly differed from all other treatments

4.3.3. Disease severity

The data on disease severity revealed that (Table 4) (Plate 10) all the treatments were significantly superior to the untreated control at 45, 60 and 75 DAS. At 45 DAS, the lowest (7.55 %) disease severity was recorded by the plant treated recorded with difenoconazole (0.1 %) with 70.62% disease suppression over the control. This was on par with propiconazole (0.1%) with a disease severity of 9.31% and 63.77% disease suppression over the control. Among the treatments, *P. fluorescens* (2%) recorded the highest (18.42%) disease severity and 28.32% disease suppression over the control. The maximum (25.70%) disease severity was recorded with untreated control plants and differed significantly from all other treatments

Disease severity recorded at 60 DAS revealed that severity of leaf spot in treated plants ranged from 8.033 to 20.96 %. Difenoconazole (0.1%)) recorded

Table 4. Effect of fungicides, organic formulations and *P. fluorescens* in leaf spot disease severity of cowpea under pot culture studies

	Mean disease severity (%)				Percentage disease suppression over control		
Trt no	Treatments	45 DAS	60 DAS	75 DAS	45 DAS	60 DAS	75 DAS
T1	Propiconazole (0.1%)	9.31 (17.76)^{dc}	10.26 (18.68)^{ef}	11.13 (19.49)de	63.77	64.7	67.62
T2	Azoxystrobin (0.1%)	18.09 (25.15)^{b}	20.96 (27.21)^{b}	26.00 (30.63)^{b}	29.61	27.97	24.37
T3	Hexaconazole (0.1%)	11.74 (20.03)^{cd}	12.36 (20.58)^{de}	13.56 (21.60)^{cd}	54.31	57.51	60.55
T4	Difenoconazole (0.1%)	7.55 (15.95)^{e}	8.033 (16.46)f	8.23 (16.67)^{e}	70.62	72.38	76.06
T5	Carbendazim (0.1%)	13.71 (21.71)^{bc}	14.35 (22.25)^{cd}	14.95 (22.73)^{bc}	46.65	50.67	56.51
T6	Neem oil (0.5%)	18.02 (24.93)^{b}	17.41 (24.63)^{bc}	21.84 (27.70)^{b}	29.88	40.15	51.29
T7	Mineral oil (0.1%)	14.29 (22.17)^{bc}	14.05 (22.00)^{cd}	16.54 (23.97)^{bc}	44.39	51.70	51.89
T8	*Pseudomonas fluorescens* (2%)	18.42 (25.24)^{b}	19.00 (25.74)^{b}	24.33 (28.63)^{b}	28.32	34.68	29.23
T9	Control	25.70 (30.45)^{a}	29.09 (32.63)^{a}	34.38 (35.87)a			
	CD(0.05)	3.871	2.672	3.871			

Mean of three replications, values in the parenthesis are arc sin transformed.
Treatments with same alphabets in the superscript do not differ significantly

the lowest 8.03% disease severity and 72.38% disease suppression over control which was on par with propiconazole (0.1%) with a disease severity of 10.26% and 64.7% disease suppression over control. Among the treatments, *P. fluorescens* (2%) registered the highest (19.00%) disease severity and 34.68% disease suppression over the control. The untreated control plants recorded maximum (29.09%) disease severity and differed significantly from all other treatments.

At 75 DAS, the plants treated with difenoconazole (0.1%) registered the lowest (8.23%) disease severity and 76.06% disease suppression over control which was on par propiconazole (0.1%) recording a disease severity (11.13%) and 67.62% disease suppression over control. Among the fungicides, the plants treated with azoxystrobin (0.1%) recorded the highest (26.00%) disease severity and 24.37% disease suppression over control which was on par with carbendazim (0.1%) with a disease severity of 14.95% and 56.51% disease suppression over control.

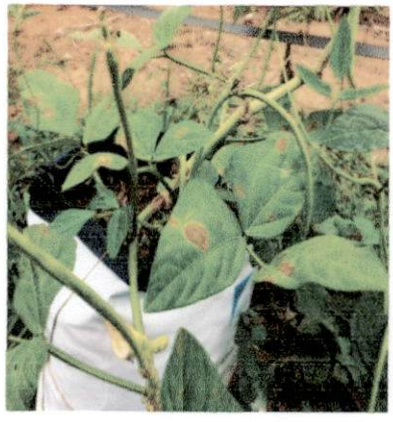
Among organic formulations, mineral oil (0.1%) recorded lowest 16.54% disease severity and 51.89% disease suppression over control which was on par with neem oil (0.5%) with a disease severity of 21.84% and 51.19% disease suppression over control.

Bio-control agent, *P. fluorescens* (2.00%) recorded disease severity of 24. 33% with disease suppression of 29.23 % over control. However, the untreated control plants recorded the maximum disease severity of 34.38 % and differed significantly from all other treatments.

4.3.4. Yield characters

Observations on yield and yield parameter such as pod number and fruit yield were recorded.

Difenoconazole (0.1%)



Propiconazole 0.1%

Mineral oil (0.1%)

Neem oil (0.5%)

Pseudomonas fluorescens 2%

Control

Plate 10. ***In vivo*** **effect of selected fungicides, organic formulations and** ***P. fluorescens*** **against** ***Cercospora cruenta***

4.3.4.1. *Pod Number*

The plants treated with difenoconazole 0.1% recorded the highest (83.66) number of pods and differed significantly from all other treatments .This was followed by treatment with propiconazole (0.1%) having 76.33 pods /pot. Among the treated plants, azoxystrobin 0.1% and neem oil (0.5%) registered the least number (55.33) of pods /pot. The lowest number of (47.33) pods/pot was noticed in untreated control plants. (Table 5)

4.3.4.2. *Pod Yield Kg/Pot*

With regard to pod yield plants treated with difenoconazole (0.1%) registered the maximum yield (0.523 kg/pot) and differed significantly from all other treatments followed y treatment with propiconazole (0.1%) (0.473 kg/pot). Among the treatments, plants treated with neem oil (0.5%) recorded the minimum yield (0.283 kg/pot). The untreated control plants registered the lowest yield (0.277kg/pot) and differed significantly from all other treatments (Table 6)

4.4. Field evaluation of selected fungicides, organic formulations and *P. fluorescens* for suppression of Cercospora leaf spot

Based on the result of pot experiment, a field study was conducted at Coconut Research Station, Balaramapuram to evaluate the efficacy of the two best fungicides *viz.*, difenoconazole (0.1%) and propiconazole (0.1%), two best organic formulations *viz.*, mineral oil (0.1% and neem oil (0.5%) and *P. fluorescens* (2.00%) (Plate 11). Observations on the number of days for the appearance of the leaf spot disease, disease incidence, disease severity and yield characters were recorded.

4.4.1. Number of days for the appearance of the leaf spot disease

The number of days taken for the first appearance of Cercospora leaf spot naturally in the field ranged from 35 days (Plate 12).

Table 5. Effect of fungicides, organic formulations and *P. fluorescens* in pod number of vegetable cowpea under pot culture studies

	Treatments	Pod number/pot	Percentage pod number increase over control
T1	Propiconazole (0.1%)	76.33^{b}	61.27
T2	Azoxystrobin (0.1%)	55.33^{de}	16.90
T3	Hexaconazole (0.1%)	70.00^{c}	47.89
T4	Difenoconazole (0.1%)	83.66^{a}	76.75
T5	Carbendazim (0.1%)	65.66^{cd}	38.72
T6	Neem oil (0.5%)	55.33^{e}	16.90
T7	Mineral oil (0.1%)	60.33^{cd}	27.46
T8	*Pseudomonas fluorescens* (2%)	63.66^{de}	34.50
T9	Control	47.33^{de}	
	CD(0.05)	7.746	

*mean of three replications. Treatments with same alphabets in the superscript do not differ significantly

Table 6. Effect of fungicides, organic formulations and *P. fluorescens* in the yield of vegetable cowpea under pot culture studies

Treatment No	Treatments	*Yield (Kg/pot)	Percentage yield increase over control
T1	Propiconazole (0.1%)	0.473 [ab]	70.76
T2	Azoxystrobin (0.1%)	0.373 [c]	34.65
T3	Hexaconazole (0. 1%)	0.453 [b]	63.53
T4	Difenoconazole (0.1%)	0.523[a]	88.80
T5	Carbendazim (0. 1%)	0.456[b]	64.62
T6	Neem oil (0.5%)	0.283[c]	2.16
T7	Mineral oil (0. 1%)	0.416[b]	50.18
T8	*Pseudomonas fluorescens* (2%)	0.426 [b]	53.79
T9	Control	0.277[d]	
	CD(0.05)	0.119	

*mean of three replications. Treatments with same alphabets in the superscript do not differ significantly

Plate 11. Field view of experimental plot

Plate 12. First appearance of Cercospora leaf spot

4.4.2. Disease incidence (%)

All the treatments were effective in the suppression of Cercospora leaf spot of cowpea on field evaluation. However, the statistical analysis of the data revealed that foliar application of difenoconazole (0.1%) registered the minimum (31.88%) disease incidence which was on par with propiconazole(0.1%)(34.78%).This was followed by carbendazim 0.1% with a disease incidence of 39.12% which was on par with mineral oil (0.1%)(40.57%).The untreated control plants recorded the highest (55.06%) disease incidence which was on par with *P. fluorescens* (2.00%) *(*47.82%) and neem oil (0.5%) (52.17 %) (Table 7).

4.4.3. Disease severity

Significant difference was noted among the treatments with respect to disease severity of Cercospora leaf spot at 45, 60 and 75 DAS. At 45DAS ,the plants treated with difenoconazole(0.1%) recorded the lowest (1.21%) disease severity and 83.21% disease suppression over the control which was on par with propiconazole (0.1%)(1.87%).The untreated control plants registered the highest (7.21%) among all the treatments

The data on disease severity at 60 DAS revealed that lowest (3.88%) disease severity was recorded with the foliar spray of difenoconazole (0.1 %) followed by propiconazole (0.1%) (5.18%) having 69.47% and 59.24% disease suppression over control. The highest (12.71%) disease severity was noticed among the untreated control plants which was on par with neem oil (0.5%) having a disease severity of 21.47% and 28.24% disease suppression over the control

Disease severity of Cercospora leaf spot recorded at 75 DAS revealed that the most effective fungicide in controlling the disease was with foliar application of difenoconazole (0.1%) having 13.97% disease severity and 63.59 per cent of disease suppression over control. This was on par with plants treated with propiconazole (0.1%) (16.39%) mineral oil (0.1%) (14.32%) and carbendazim

Table 7. Effect of fungicides, organic formulations and *P fluorescens* in leaf spot disease incidence of vegetable cowpea in field condition studies

Treatment No	Treatments	*Mean disease incidence (%) 75DAS	Percentage reduction of disease incidence over control
T1	Propiconazole (0.1%)	34.78 (35.34) ^{cd}	36.83
T2	Difenoconazole (0.1%)	31.88 (34.36)^{cd}	42.10
T3	Neem oil (0.5%)	52.17 (46.53) ^{ab}	5.24
T4	Mineral oil (0.1%)	40.57 (39.70) ^{bcd}	26.31
T5	*Pseudomonas fluorescens (2%)*	47.82 (43.10) ^{abc}	13.15
T6	Chemical control check Carbendazim (0.1%)	39.12 (39.02) ^{bcd}	28.95
T7	Control	55.06 (49.22) ^{a}	
	CD(0.05)	13.275	

Mean of three replications, values in the parenthesis are arc sin transformed. Treatments with same alphabets in the superscript do not differ significantly.

Table 8. Effect of fungicides, organic formulations and *P . fluorescens* on Cercospora leaf spot disease severity of cowpea in field condition

		* Percentage disease index (%)			Percentage disease suppression over control		
	Treatments	45 DAS	60 DAS	75DAS	45 DAS	60 DAS	75 DAS
T1	Propiconazole (0.1%)	1.87 (7.64))^{cd}	5.18 (8.16)^{c}	16.39 (23.85) ^{bc}	74.06	59.24	57.28
T2	Difenconazole (0.1%)	1.21 (6.23) ^{d}	3.88 (7.34) ^{c}	13.97 (21.85) ^{c}	83.21	69.47	63.59
T3	Neem oil (0.5%)	5.85 (13.80) ^{ab}	9.12 (13.90)^{ab}	21.47 (27.55) ^{b}	18.86	28.24	43.93
T4	Mineral oil (0.1%)	5.07 (12.92) ^{ab}	10.80 (10.60) ^{bc}	14.32 (22.21) ^{c}	29.68	15.02	62.67
T5	*Pseudomonas fluorescens* (2%)	3.27 (10.38) ^{bc}	10.50 (8.81) ^{c}	21.76 (27.68) ^{b}	54.64	17.38	43.28
T6	Chemical control check Carbendazim (0.1%)	3.40 (10.60) ^{bc}	7.11 (10.50) ^{bc}	17.13 (24.37) ^{bc}	52.84	44.05	55.35
T7	Control	7.21 (15.51) ^{a}	12.71 (15.51) ^{a}	38.37 (38.27) ^{a}			
	CD(0.05)	3.523	3.593	4.13			

Mean of three replications, values in the parenthesis are arc sin transformed.

Treatments with same alphabets in the superscript do not differ significantly.

Difenoconazole (0.1%)

Propiconazole (0.1%)

Mineral oil (0.1%)

Neem oil (0.5%)

P. fluorescens (2%)

Control

Plate 13. Effect of fungicides, organic formulations and *P. fluorescens* aganist *Cercospora cruenta* under field conditions

(0.1%) (17.13%).The bio control agent *P. fluorescens* (2%) registered a disease severity of 21.76% which was on par with the neem oil 0.5% (21.47%) having 43.28 and 44.04% disease suppression over the control . Among all the treatments the untreated control plants recorded the highest (38.37%) disease severity and differed significantly from all other treatments (Table 8) (Plate 13)

4.4.4. Yield characters

Observations on yield and yield parameter such as pod number and fruit yield were recorded.

4.4.4.1. *Pod Number*

The statistical analysis on pod number per plot revealed that the maximum number of pods (360.00 pods/ plot) was observed with 0.1 per cent difenoconazole and differed significantly from all other treatments with 99.26% increase of pod number over control. This was followed by propiconazole (0.1%) (343pods/plot) with 60.11% increase of pod number over control. Among the organic formulations, mineral oil (0.1%) recorded the maximum (304/plot) number of pods which was on par with neem oil (0.5%) (277.66 pods/plot). The KAU talc based formulation of *P. fluorescens* recorded pod number of 313.66 /plot with 34.50 per cent increase of pod number over control. However, the untreated control plants recorded the least (180.66) number of pods /plot (Table 9).

4.4.4.2. *Pod yield kg/pot*

Data on cowpea pod yield (kg/ha) as influenced by the different treatments are furnished in table 10.

Analysis of data on pod yield indicated that plants treated with as difenoconazole (0.1%) registered the maximum (1620.0 kg/ha) yield with 87.71% increase over the control and differed significantly from all other treatments. This was followed by propiconazole (0.1%) with 1386.5 kg/ha pod yield and 60.60 %

Table 9 . Effect of fungicides, organic formulations and *P. fluorescens* in pod number of vegetable cowpea in field studies

Treatment No	Treatments	Pod number /plant	Pod number/ Plot $20m^2$	Percentage pod number increase over control
T1	Propiconazole (0.1%)	14.91	343.00[b]	89.85
T2	Difenoconazole (0.1%)	15.65	360.00[a]	99.26
T3	Neem oil (0.5%)	12.07	277.66[bc]	53.69
T4	Mineral oil (0.1%)	13.21	304.00[bc]	68.27
T5	Carbendazim (0.1%)	13.63	334.00[b]	84.87
T6	*Pseudomonas fluorescens* (2%)	14.52	313.66[c]	73.25
T7	Control	7.85	180.66[b]	
	CD(0.05)		0.842	

Mean of three replications. Treatment with same alphabets in the superscript do not differ significantly

Table 10. Effect of fungicides, organic formulations and *P. fluorescens* in the yield of vegetable cowpea under field studies

Treatment No	Treatments	Yield (kg/plot) ($20\ m^2$)	Yield (kg/ha)	Yield increase over control
T1	Propiconazole (0.1%)	2.773^{ab}	1386.5	60.66
T2	Difenoconazole (0.1%)	3.240^{a}	1620.0	87.71
T3	Neem oil (0.5%)	2.193^{cd}	1096.5	27.06
T4	Mineral oil (0.1%)	2.586^{b}	1293.0	49.83
T5	*Pseudomonas fluorescens* (2%)	2.636^{b}	1318.0	52.27
T6	Chemical control check Carbendazim (0 .1%)	2.910^{b}	1455.0	68.59
T7	Control	1.726^{c}	863.0	
CD(0.05)		0.842		

Mean of three replications. Treatments with same alphabets in the superscript do not differ significantly.

Table 11. Economic analysis of the experimental entitled management of Cercospora leaf spot of vegetable cowpea *Vigna unguiculata* subsp *sesquipedalis* (L)Verdcourt under field condition

Treatment No	Treatments	Yield (kg/ha)	Additional yield over control	Additional Income (Rs/ha)	Additional Expenses (Rs/ha)	Benefit: Cost ratio
T1	Propiconazole (0.1%)	1386.5	523.5	31,410	13,965	2.25
T2	Difenoconazole (0.1%)	1620.0	757.0	45,420	18,600	2.44
T3	Neem oil (0.5%)	1096.5	233.5	14,010	11,850	1.18
T4	Mineral oil (0.1%)	1293.0	430.0	25,800	11,700	2.21
T5	*Pseudomonas fluorescens* (2%)	1318.0	455.0	27,300	13,500	2.02
T6	Chemical control check Carbendazim (0 .1%)	1355.0	492.0	29,520	12,405	2.37
T7	Control	863.0				

pod yield increase over the control. Among the organic formulations, mineral oil (0.1%) treated plants recorded the highest pod yield of 1293 kg/ha which was on par with neem oil (0.5%) which yielded 1096.5 kg/ha having 49.83% and 27.06% yield increase over control. Foliar spray of KAU talc based formulation of *P. fluorescens* (2.00%) recorded pod yield of 1318 kg/ha with 52.27% increase of pod yield over control. The untreated control plants recorded the least (863kg/ha) among all the treatments.

4.4.5. Benefit : Cost Ratio

The data on benefit:cost ratio of field evaluation of fungicides, organic formulations and *P. fluorescens* revealed that the returns from treatment with difenoconazole (0.1% recorded the highest (2.44) returns among all the treatments and the lowest(1.18) returns was recorded from neem oil(0.5%)(Table 11).

4.4.6. Occurrence of other pests and diseases

Minor incidence of mosaic disease was observed in the field. Among the insect pest, infestation of aphids and pod bugs were observed.

DISCUSSION

5. DISCUSSION

Cowpea is the most important legume vegetable grown in Kerala. Cercospora leaf spots, affecting the leaves and stem occurring at all stages of the crop are at present causing serious economic loss to the crop through severe defoliation. The fungus has a wide host range of cultivated and wild plants. The continuous cultivation of cowpea in one or the other area as well as wild/weed hosts enable the fungus to survive and disseminate conidia by wind. There have been no detailed cultural / morphological studies on the fungal pathogen, epidemiology and potential of Cercospora leaf spot disease (CLS) in cowpea in Kerala. In this context the present study was undertaken to study the symptomatology and etiology of the pathogen ,efficacy of fungicides organic formulations, and bio-control agent on the infection, disease development, disease incidence and yield of cowpea under pot culture and field conditions.

Symptomatology of the Cercospora leaf spot was studied by observing the infected samples from the field near College of Agriculture,Vellayani. The symptoms were observed on the leaves, pods and stem. The characteristics symptoms of *Pseudoercospora cruenta* on leaves appeared initially as small light greenish yellow diffused spots on the adaxial surface of the leaf which later became reddish brown circular to angular, 2-10 mm diameter with a greenish yellow halo with black mass of conidia on the upper and lower surfaces . At the final stage, the leaves became necrotic, dried up and hung down. On the stem, symptom appeared as black coloured lesions with mass of conidia. In pods, irregular black spots appeared with powdery mass of fungal growth on and inside the fruits, damaging them completely. The symptoms observed were similar to the depictions given by Vakli, 1977 and Emechebe and Shoyinka, 1985.

Symptoms of *Cercospora canescens* on leaves appeared as rough, diffused and irregular, dark red spots with a silvery grey centre and a darker margin. Conidia were produced on the corresponding lower surface of the leaves.

Vakli (1977) also described the symptoms of Cercospora leaf spot of cowpea caused by *C. canescens* on leaves as round orange to light brown, silvery grey spots of 8-15 mm diameter. On the petiole, symptom appeared as uniform light brown spindle shaped lesion.

Conidial characters such as conidial length, width, colour and septation were recorded by microscopic studies. The results revealed that conidia produced from the diseased leaf samples were hyaline, obclavate, straight to curved tip and 10 to17 septate with conidial length ranging from 24.7 to 57.6 µm and width of 1.1to 2.7µm. Similar observations were made by Chupp (1954) who reported that the conidia of *C. cruenta* were hyaline to sub-hyaline, olivaceous brown, slightly curved to straight tip with acute ends, 5-9 septate. The average size of the conidia was maximum (57.6 x 2.7 µm) for the diseased leaf samples collected from Balaramapuram followed by Kakkamoola (55. 2 x 2.1 µm) . The minimum average size was recorded with leaf samples collected from Poonkulam (24.64 x 1.74µm). The results were in accordance with the findings of Ellis (1976) who described that the conidial size of *C cruenta* ranged from 40-150 x 3-4.5 µm. Vasudeva (1963) also noticed that the conidial size of *C cruenta* in cowpea ranged from 60-80 x 4 µm

Mycelia of the pathogen was very fine to coarse, fairly regular to very irregular, well developed, branched, intercellular with septate hyphae. Similar observations were made by Solheim (1929) who reported that the the mycelium of *Cercospora* appeared to be very fine to coarse, fairly regular to very irregular, septate hyphae which for the most part ramify the host tissue.

Based on the conidial and mycelial characters, the pathogen was tentatively identified as *Cercospora* sp. The identity of the pathogen was further confirmed based on the morphological characteristics and the herbarium specimen send to National Fungal Culture Collection of India, (N.F.C.C.I), Pune as *Pseudocercospora* sp. aff. *Pseudocercospora vignigena* (F: Mycosphaerellaceae)

The results of *in vitro* evaluation of fungicides, organic formulations and *P. fluorescens* on the conidial germination of *Cercospora* sp revealed that all the fungicides tested *in vitro* aganist *Cercospora* sp significantly inhibited the conidial germination of pathogen at two concentration over the control. Of the fungicides tested, difenoconazole (0.1%) recorded the lowest percentage (53.87 %) of conidial germination with 42.96% reduction over the control. This was followed by propiconazole (0.1%) with 59.63% of conidial germination. Efficacy of difenoconazole and propiconazole in the inhibition of the spore germination of *Cercospora* has been reported earlier (Dahmen *et al.* (1992) and Hemachandra (2007).The inhibition of spore germination was due to the inhibition of cytochrome P-45 enzyme affecting C-14 demethylation which was essential for the development of the fungal cell membrane ergosterol (Dahmen *et al.*,1992). Swamy (2010) also reported the complete inhibition of mycelial growth of *C. capsici* infecting chili under *in vitro* condition with propiconazole .Conidial germination was recorded highest (83.66%) in copper hydroxide (0.1%) with 11.41% reduction over the control.

In the case of organic formulations, mineral oil (0.1%) and neem oil (0.5%) recorded 73.77% conidial germination with 21.88 % inhibition of conidial germination over control. The findings were in agreement with Benagi (1995) and Khadar (1999) who reported the efficacy of leaf extracts of *A. indica* and neem seed kernel extract (NSKE) against conidial germination of *C. personata* and *C. arachidicola* respectively.

The recommended dosage of the KAU talc based formulation of *P. fluorescens at* 2.00% recorded only 11.88 % reduction of conidial germination over control .Simonetti (2012) also noted the *in vitro* efficacy of *P.fluorescens* against leaf spot of soybean caused by *C. sojina* and found complete inhibition of the mycelial growth and spore germination of the pathogen.

A pot culture study was conducted at Coconut Research Station, Balaramapuram to evaluate the five best fungicides (difenoconazole (0.1%), propiconazole (0.1%), azoxystrobin (0.1%), carbendazim (0.1%), hexaconazole (0.1%), two best organic formulations (mineral oil (0.1%), neem oil (0.5%) and *P. fluorescens* (2%) on Cercospora leaf spot management selected from *in vitro* evaluation. On artificial inoculation with the infected leaf samples Cercospora leaf spot appeared after 15 to 20 days in all the plants.

The results of *in vivo* evaluation revealed that, of the fungicides tested at 45DAS difenoconazole (0.1%) recorded the lowest (17.16 %) disease incidence with 64.49 % disease suppression over the control followed by propiconazole (0.1%) with 23.32% disease incidence and hexaconazole (0.1%) with 51.74 % disease incidence. Similarly, the disease incidence estimated at 60 DAS revealed that the least incidence was recorded with difenoconazole (0.1%) and propiconazole (0.1%) sprayed plants with 54.10 % and 52.82% disease incidence respectively followed by hexaconazole (0.1%) and carbendazim (0.1%) with 49.54% and 43.45% disease incidence respectively. At 75 DAS, among the fungicides the lowest (23.80%) disease incidence was recorded with difenoconazole (0.1%) with 54.80% of disease suppression over the control followed by propiconazole 0.1% (26.83%),hexaconazole(0.1%),(27.33%) and carbendazim (0.1%)(31.09%).

The findings were in agreement with the results of Bhat *et al.* (2015) who reported that the foliar spray of carbendazim (0.05%) and difenoconazole(0.02 %) significantly reduced the pod infection (77 %) and leaf spot intensity (68 %) of *C. canescens* of green gram. Similarly, Kapadiya and Dhruji (1999) also reported that the minimum mean disease intensity (36.4 %) and maximum disease control (61.01 %) was recorded with the foliar application of 0.0125 per cent difenoconazole followed by propiconazole, hexaconazole, carbendazim and mancozeb in cowpea aganist Cercospora leaf spot. Similar observations had been made by many workers as well(West,1983; Das and Mohanty, 1986 ;Vaishnav *et*

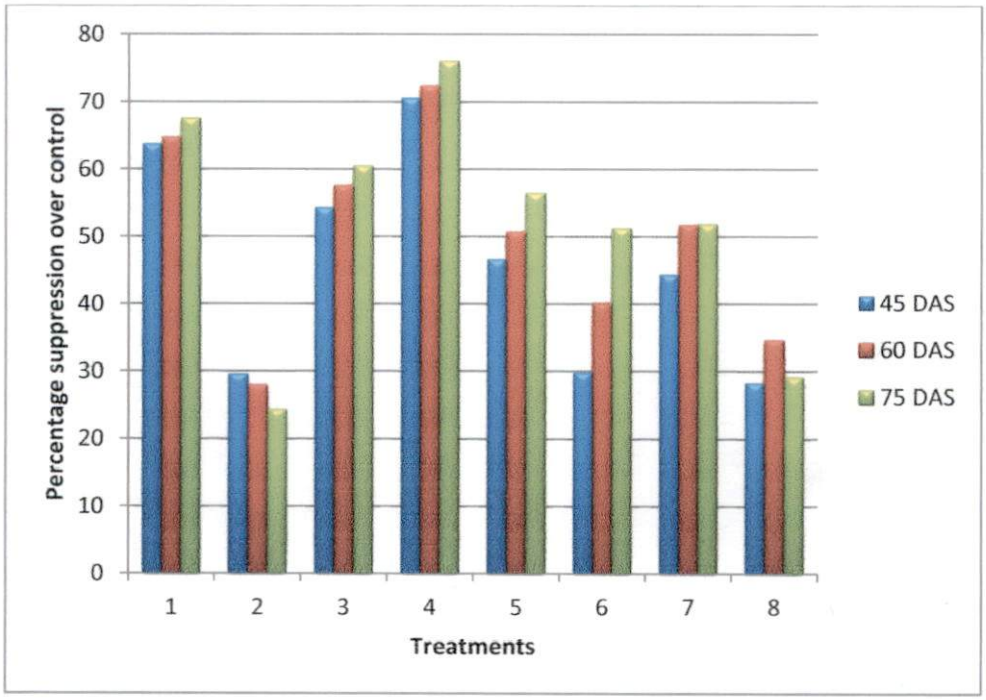


Fig 1.Effect of fungicides, organic formulations and *P fluorescens* on Cercospora leaf spot disease severity in cowpea under field condition

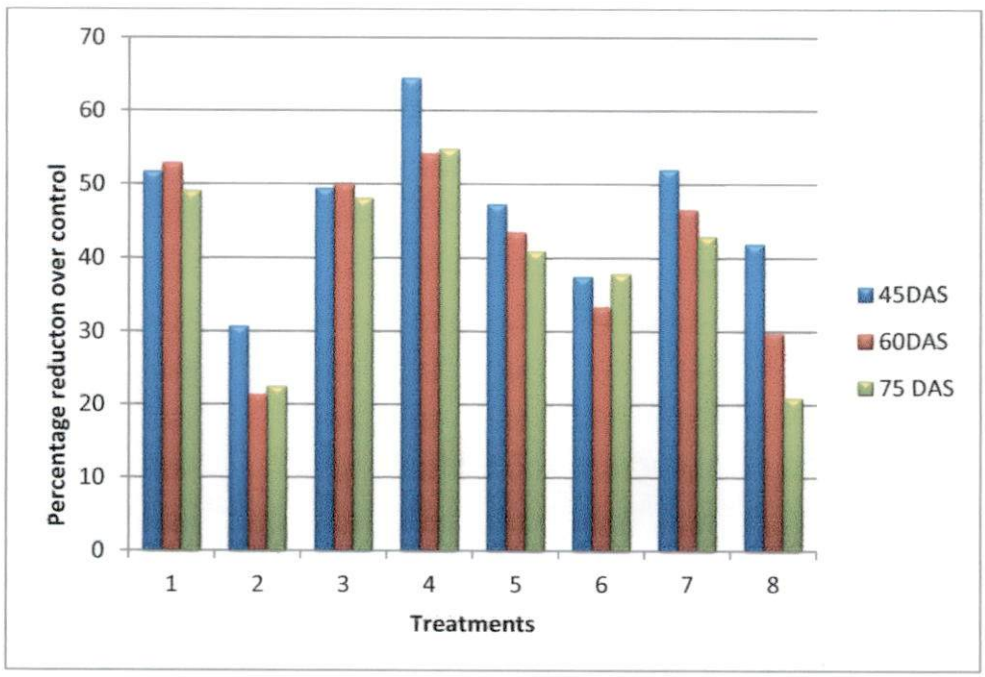


Fig: 2..Effect of fungicides, organic formulations and *P fluorescens* on Cercospora leaf spot disease incidence in cowpea under pot culture experiment

al. (1992) ;Tiwari and Kotasthane, 1984 Kaur *et al.*(2004); Saxena and Tripathi, 2006) ;Hundekar *et al.*(2005); Jahagirdar and Hundekar, 2010).

At 45 DAS, among the treatments, highest (33.47%) disease incidence was observed in azoxystrobin (0.1%) with only 30.74 % disease suppression over the control. At 60 and 75DAS also disease incidence was recorded highest in azoxystrobin (0.1%) with 38.04%) and (40.83%) respectively.

At 75 DAS, among the organic formulations mineral oil (0.1 %) showed the lowest (30.04%) disease incidence with 42.95% disease suppression over the control and followed by with neem oil (0.5%) with 37.9 % disease suppression over control. The results were in accordance with Trivedi (2014) who reported the efficacy of neem oil aganist the Cercospora leaf spot in black gram caused by *C. canescens.* Srivastava and Nelson (2012) also reported that neem oil (70 %) was found effective against cercospora leaf spot of egg plant.

At 75 DAS, among all the treatments, *P. fluorescens* recorded the highest (41.64%) disease incidence with only 20.92% disease suppression over the control. However, Pairashi *et al.* (2007) noticed the reduction in the incidence of leaf spot in tobacco caused by *C. nicotiane* by the application of *P. fluorescens* at the rate of 2 g/L.

The data on disease severity revealed that at 45 DAS, the lowest (7.55 %) disease severity was recorded by the plant treated with difenoconazole (0.1 %) with 70.62% disease suppression over the control followed by propiconazole (0.1%) with a disease severity of 9.31% and 63.77% disease suppression over the control. Disease severity recorded at 60 DAS revealed that difenoconazole (0.1%)) recorded the lowest (8.03%) disease severity and 72.38% disease suppression over control followed by propiconazole (0.1%) with a disease severity of 10.26% and 64.7% disease suppression over control. At 75 DAS, the plants treated with difenoconazole (0.1%) registered the lowest (8.23%) disease severity and 76.06% disease suppression over control followed by propiconazole (0.1%) recording a disease severity (11.13%) and 67.62% disease suppression

over control. Similar observations were made by Sunkad and Mesta (2005) reported that propiconazole, hexaconazole and difenoconazole at 0.1 per cent recorded the lowest per cent disease index of 71.59 % 66.93%, 65. 21% respectively against cercospora leaf spot in groundnut .Swamy (2010) also recorded that the fungicides propiconazole and carbendazim at 0.1% sprayed for two times at 10 days interval were most effective in reducing the Cercospora leaf spot of chilli with per cent disease index of 11.67 and 16.48 respectively.

Among the fungicides, the plants treated with azoxystrobin (0.1%) recorded the highest (26.00%) disease severity and 24.37% disease suppression over control with carbendazim (0.1%) with a disease severity of 14.95% and 56.51% disease suppression over control.

Among organic formulations, mineral oil (0.1%) recorded lowest 16.54% disease severity and 51.89% disease suppression over control followed by neem oil (0.5%) with a disease severity of 21.84% and 51.19% disease suppression over control. Under field condition, foliar spray of neem oil at 0.2% recorded the lowest percentage disease index of 29.38%. Earlier works conducted by Kumar *et al.* (2017) foliar spray of neem oil at 0.2% recorded the lowest percentage disease index of 29.38% against Tikka leaf spot of groundnut caused by *Cercospora* sp.

However, bio-control agent, *P. fluorescens* (2.00%) recorded much higher disease severity of 24. 33% with only 29.23 % of disease suppression over control. On the contrarary, Poornima (2010) reported that *P. fluorescens* (0.2%) gave a good result with less percent disease index (4.16) of leaf spot of palak. Singh *et al.* (2012) also reported that less percent disease control (PDC) due to the use of *P. fluorescens* was obtained against Cercospora leaf spots of cotton which ranged from 34.7 to 47.7 per cent.

With respect to yield parameters, the plants treated with difenoconazole 0.1% recorded the highest(83.66) number of pods while, plants treated, with azoxystrobin 0.1% and neem oil (0.5%) registered the least number(55.33)of pods /pot. The lowest number of (47.33) pods/pot was noticed in untreated control

plant. With regard to pod yield plants treated with difenoconazole (0.1%) registered the maximum yield (0.523 kg/pot) followed treatment with propiconazole (0.1%) (0.473 kg/ pot). Among the treatments, plants treated with neem oil (0.5%) recorded the minimum yield (0.283kg/pot).The untreated control plants registered the lowest yield (0.277kg/pot) and differed significantly from all other treatments. Studies conducted by Veena *et al.* (2014) reported that foliar application of difenoconazole (0.1%) gave superior pod number and fruit yield when compared to untreated control in green gram. Similarly, Bhat *et al.* (2015) reported that the foliar spray of carbendazim (0.05%) and difenoconazole(0.02 %) and gave an attractive seed yield of 8.62-8.95 q/ha .

Based on the result of pot experiment, a field study was conducted at Coconut Research Station, Balaramapuram to evaluate the efficacy of the two best fungicides *viz.*, difenoconazole (0.1%) and propiconazole (0.1%), two best organic formulations *viz.*, mineral oil (0.1% and neem oil (0.5%) and *P. fluorescens* (2.00%). Observations on the number of days for the appearance of the leaf spot disease, disease incidence, disease severity and yield characters were recorded.

The number of days taken for the first appearance of Cercospora leaf spot naturally in the field ranged from 30 to 40 days. Cercospora leaf spot symptoms usually appear first on cowpea leaves within four weeks after planting coinciding with the oneset of flowering. (Williams,1975). Schneider and Scinclair (1975) observed that some fungitoxic compound were associated with the younger leaves of the susceptible cultivar and henceforth the older leaves were severely diseased and the plants in the pre flowering stage were free of disease.

With regards to disease incidence, all the treatments were found effective in the suppression of Cercospora leaf spot of cowpea on field evaluation at 65DAS. However, foliar application of difenoconazole (0.1%) registered the minimum (31.88%) disease incidence followed by propiconazole(0.1%)(34.78%) and carbendazim 0,1% with a disease incidence of 39.12%. The data observed

was in accordance with Kapadiya and Dhruji, (1999) who reported that .minimum mean disease intensity (36.4 %) and maximum disease control (61.01 %) was recorded with the foliar application of 0.0125 per cent difenoconazole followed by propiconazole, hexaconazole, carbendazim and in cowpea aganist Cercospora leaf spot. Similar observations had been made by other workers as well(West (1983; Das and Mohanty 1986 Vaishnav *et al.* (1992) Tiwari and Kotasthane, 1984 Kaur *et al.*, 2004 Saxena and Tripathi, 2006) (Hundekar *et al.,* 2005; Jahagirdar and Hundekar, 2010).

The untreated control plants recorded the highest (55.06%) disease incidence which was on par with *P. fluorescens* (2.00%) (47.82%) and neem oil (0.5%) (52.17 %).However, Raguchander *et al.* (2005) reported the foliar spray of *P. fluorescens* (2.00%) reduced the Cercospora leaf spot disease incidence by 94.8% in Urd bean.

The data on disease severity at 45 DAS revealed that, the plants treated with difenoconazole(0.1%) recorded the lowest (1.21%) disease severity and 74.73% disease suppression over the control which was followed by propiconazole (0.1%)(1.87%). The untreated control plants registered the highest (7.21%) among all the treatments. At 60 DAS, the lowest (3.88%) disease severity was recorded with the foliar spray of difenoconazole (0.1 %) followed by propiconazole(0.1%) (5.18%) having 63.59% and 57.28% disease suppression over control. At 75 DAS, it was noticed that the most effective fungicide in controlling the Cercospora leaf spot was with foliar application of difenoconazole (0.1%) having 13.97% disease severity and 63.59 per cent of disease suppression over control followed by propiconazole (0.1%) (16.39%) and carbendazim (0.1%) (17.13%). Sunkad and Mishra, (2005)also reported that the triazole fungicides such as propiconazole, hexaconazole and difenoconazole at 0.1 per cent recorded the lowest per cent disease index of 71.59 % 66.93%, 65. 21% respectively against cercospora leaf spot in groundnut.

In the case of organic formulations, mineral oil recorded a disease severity of 14.32% with 62.67% reduction over the control followed by neem oil (0.5%) with a disease severity of 21.47%. Studies conducted by Stover (1990) revealed that, three sprays of propiconazole 0.05%+ petroleum based mineral oil 1% effectively controlled (13.7 8 %) the Sigatoka leaf spot disease in Banana. The activity of this group of oils had been shown to delay symptom development and inhibit lesion growth by reducing spore germination, germ-tube growth and sporulation of the Sigatoka pathogen. Trivedi (2014) also reported that under field conditions, foliar application of neem oil at 0.2% recorded the lowest percentage disease index of 29.38% against the Cercospora leaf spot in black gram caused by *C. canescens*.

The bio control agent *P. fluorescens* (2%) registered a disease severity of 21.76% having 43.28 % disease suppression over the control. Devappa and Thejakumar (2016) reported that the foliar application of propiconazole @ 0.05% + *P. fluorescens*@ 5g/L effectively managed the Cercospora leaf spot disease with a Percent disease index ranging from 10.67 to 25.33 compared to control.

Application of triazole fungicides reported uniform reduction of disease severity while, oil based formulations and *P. fluorescens* shows an increase during the final spray. The reason for this may be due to the differnce in their mode of action and change in climatic conditions. Lyr, (1987) reported that the triazole fungicides inhibit the specific enzyme, C-14demethylase, which plays a role in sterol production. These results are in accordance with the hypothesis that the DMI fungicides are best applied prior to infection or in the later stage for the control of leaf spot disease.

The analysis of data on pod yield parameters such as pod number per plot revealed that the maximum number of pods (360.00 pods/plot) were observed with 0.1 per cent difenoconazole with 99.26 % increase of pod number over control. This was followed by propiconazole (0.1%) (343 pods/plot) with 89.85% increase of pod number over control. With respect to pod yield, plants treated with

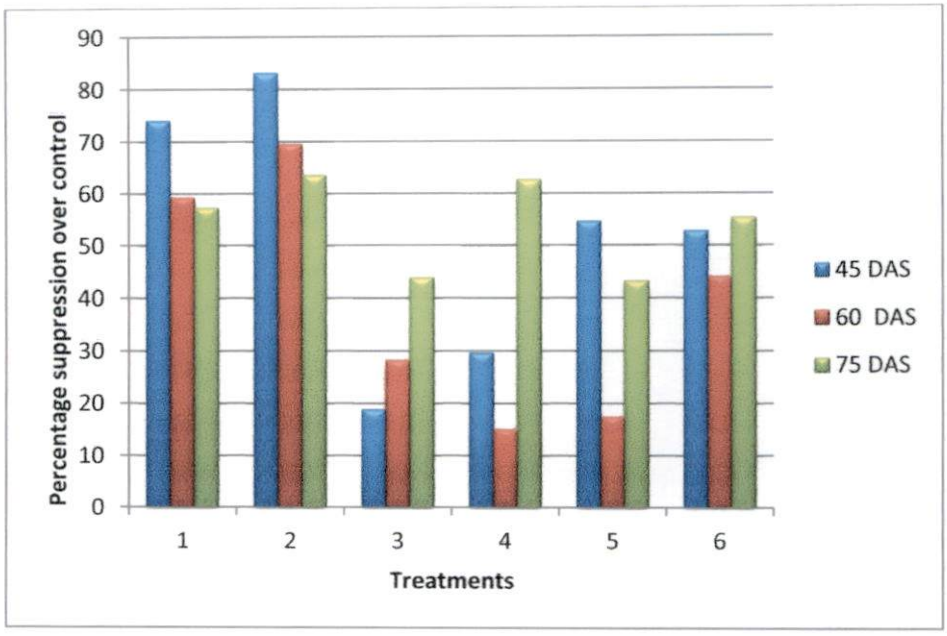


Fig 3. Effect of fungicides, organic formulations and *P fluorescens* on Cercospora leaf spot disease severity in cowpea under field condition.

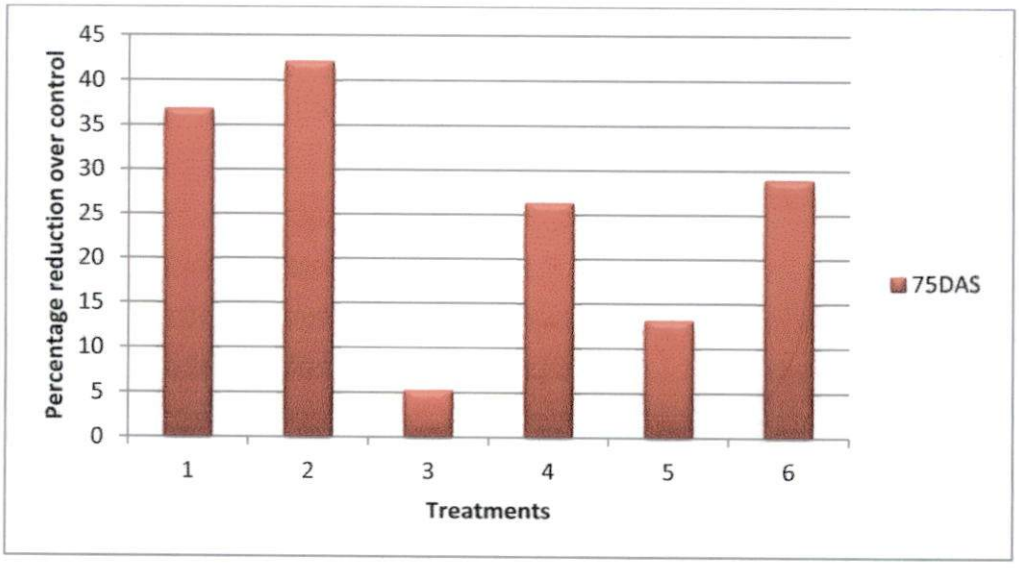


Fig 4.Effect of fungicides, organic formulations and *P fluorescens* on Cercospora leaf spot disease incidence in cowpea under pot culture experiment

as difenoconazole (0.1%) registered the maximum (1620 kg/ha) yield with 87.71% increase over the control followed by propiconazole (0.1%) with (1386.5kg/ha) pod yield and 60.66% pod yield increase over the control. Veena *et al.* (2014) reported that foliar application of 0.1 per cent difenoconazole gave superior pod number and fruit yield when compared to untreated control in green gram. Separate foliar application of propiconazole and carbendazim increased pod number and fruit yield in case of *Cercospora canescens* in green gram and black gram (Jameel *et al* .,2014)

Among the organic formulations, mineral oil (0.1%) recorded the maximum (304/plot) number of pods followed by neem oil (0.5%) (277.66 pods/plot). With regard to pod yield, mineral oil (0.1%) treated plants recorded the highest yield of 1293 kg/ha followed neem oil (0.5%) which yielded 1096.5 kg/ha having 49.83 and 27.06% yield increase over control respectively. Thammaiah (2013) reported that treatment with propiconazole 0.05% + petroleum based mineral oil 1% recorded an yield of 72.28t/ha against Cercospora leaf spot in banana .

The KAU talc based formulation of *P. fluorescens* recorded pod number of 313.66/plot with 73.25 per cent increase of pod number over control. Regarding the yield, foliar spray of *P. fluorescens* (2.00%) recorded an yield of 1318 kg/ha with 52.27% increase of pod yield over control. Similar results were obtained by Kumar *et al.* (2017) who reported that foliar application of *P. fluorescens* (2%) recorded a significant increase in the plant height, maximum number of leaves, pod yield and the minimum disease intensity (20.1%).

The data on economic analysis on the field evaluation of fungicides, organic formulations and *P. fluorescens* revealed that the returns from treatment with difenoconazole (0.1% recorded the highest (2.44:1) returns among all the treatments and the lowest(1.18:1) returns was recorded from neem oil(0.5%). Similarly, Bhat *et al.* (2015) also reported that the foliar spray of difenoconazole (0.02 %) significantly reduced the Cercospora leaf spot intensity of green gram at an acceptable B:C ratio (6.38:1).

Thus the overall results of the study indicated that foliar application of difenoconazole (0.1%) at an interval of 35, 50 and 65 DAS was found to be the most effective treatment in managing Cercospora leaf spot in vegetable cowpea besides supporting proportionately impressive pod yield (1620 kg/ha) at an acceptable B:C ratio (2.44:1).Among the organic formulations, mineral oil (0.1%) was found to be the effective treatment in controlling leaf spot with a pod yield of 1293 kg/ha . While, *P. fluorescens* recorded a pod yield of 1318 kg/ha with B: C ratio of 2:02

SUMMARY

6. SUMMARY

The study entitled "Management of Cercospora leaf spot of vegetable cowpea *Vigna unguiculata* subsp *sequipedalis* (L.) Verdcourt was undertaken at the Department of Plant Pathology, College of Agriculture Vellayani and Coconut Research Station, Balaramapuram during the year 2015-2017. The study has been conducted with the objective to study the symptomatology and etiology of Cercospora leaf spot of cowpea and to develop a management strategy.

The salient results of the study are summarized below:

Field samples of Cercospora leaf spot incidence in vegetable cowpea were collected from Balaramapuram, Vellayani, Kalliyoor, Kakkamoola, Poonkulam and Manakadu locations. Studies on the symptomatology and etiology revealed that symptoms were observed on the leaves, pods and stem of cowpea. Symptoms of *Pseudocercospora cruenta* appeared initially as small light greenish yellow diffused spots on the adaxial surface of the leaf turning reddish brown with a greenish yellow halo and black mass of conidia on the upper and lower surfaces. At the final stage, the leaves became necrotic, dried up and hung down. On the stem, symptom appeared as black coloured leisons with mass of conidia and in the pods, irregular black spots appeared with powdery mass of fungal growth. Symptoms of *Cercospora canescens* on leaves appeared as rough, diffused and irregular, dark red spots with a silvery grey centre and a darker margin.

With respect to conidial characters, the conidial length ranged from 24.7 to 57.6 µm and width of 1.1to 2.7µm. Maximum size (57.6 x 2.7 µm) of the conidia was collected from Balaramapuram while, the minimum size (24.64 x 1.74µm) was recorded with samples collected from Poonkulam. Conidial septation ranged from 10t o17 in number. Mycelia of the pathogen was very fine to coarse, fairly

regular to very irregualar, well developed, branched, intercellular with septate hyphae. Based on the conidial and mycelial characters, the pathogen was tentatively identified as *Cercospora* sp. The identity of the pathogen was further confirmed based on the morphological characteristics and the herbarium specimen send to National Fungal Culture Collection of India, (N.F.C.C.I), Pune as *Pseudocercospora* sp. aff. *Pseudocercospora vignigena* (F: Mycosphaerellaceae)

The results of efficacy of fungicides, organic formulations and *P. fluorescens* on the conidial germination of *Cercospora* sp revealed that all the fungicides tested *in vitro* aganist *Cercospora* sp significantly inhibited the conidial germination of pathogen at two concentrations over the control. Of the fungicides tested, difenoconazole (0.1%) recorded the lowest percentage (53.87 %) of conidial germination with 42.96% reduction over the control while copper hydroxide (0.1%) recorded the highest (83.66%) with 11.41% reduction over the control.

A pot culture study was conducted at Coconut Research Station, Balaramapuram to evaluate the five best fungicides *viz.,* difenoconazole (0.1%), propiconazole (0.1%), azoxystrobin (0.1%), carbendazim (0.1%), hexaconazole (0.1%), two best organic formulations *viz.,* mineral oil (0.1%), neem oil (0.5%) and *P. fluorescens* (2%) on Cercospora leaf spot management selected from *in vitro* evaluation.

On artificial inoculation with the infected leaf samples Cercospora leaf spot appeared after 15 to 20 days in all the plants. Disease incidence and disease severity were recorded from 45 to 75DAS. Results of disease incidence and disease severity recorded at 75 DAS revealed that foliar spray of difenoconazole (0.1%) recorded the minimum (23.80%) disease incidence and disease severity (8.23 %). In the case of organic formulations, mineral oil (0.1%) recorded the minimum (30.04%) disease incidence and least (16.54%) disease severity. The plants sprayed with *P. fluorescens* (2.0%) recorded disease incidence of 41.64 % and disease severity of 24.33%.

Result on yield parameters such as pod number revealed that the plants treated with difenoconazole (0.1%) recorded the highest (83.66) number of pods while, azoxystrobin (0.1%) and neem oil (0.5%) registered the least (55.33) number of pods/pot. With regard to pod yield, the plants treated with difenoconazole (0.1%) registered the maximum (0.523 kg/pot) yield with 88.80% yield increase over control while ,the untreated control plants registered the lowest yield (0.277kg/pot).

Based on the results of pot experiment, a field study was conducted at Coconut Research Station, Balaramapuram to evaluate the efficacy of the two best fungicides *viz.,* difenoconazole (0.1%) and propiconazole (0.1%), two best organic formulations *viz.*, mineral oil (0.1% and neem oil (0.5%) and *P. fluorescens* (2.00%). The number of days taken for the first appearance of Cercospora leaf spot disease naturally in field ranged from 35 to 40 days. The result of disease incidence recorded at 75DAS under field evaluation revealed that foliar application of difenoconazole (0.1%) registered the minimum (31.88%) disease incidence while, *P. fluorescens* (2.00%) (47.82%) and neem oil (0.5%) (52.17 %) recorded the highest disease incidence.

Result of disease severity recorded at 45 DAS revealed that the plants treated with difenoconazole (0.1%) recorded the lowest (1.21%) disease severity with 83.21% disease suppression over the control. The untreated control plants registered the highest (7.21%) disease severity among all the treatments. The data on disease severity recorded at 60 DAS revealed that lowest (3.88%) disease severity was with foliar spray of difenoconazole (0.1 %) while, the highest (12.71%) disease severity was noticed with neem oil (0.5%) having 28.24% disease suppression over the control.

Disease severity of Cercospora leaf spot recorded at 75 DAS revealed that, the most effective fungicide in controlling the disease was foliar application of difenoconazole (0.1%) having 13.97% disease severity with 63.59 % disease suppression over the control. Among organic formulations, mineral oil (0.1%)

recorded the lowest disease severity (14.32%) with 62.67% disease suppression over the control. The bio control agent *P. fluorescens* (2.00%) registered a disease severity of 21.76% with only 43.28% disease suppression over control. Among all the treatments the untreated control plants recorded the highest (38.37%) disease severity.

Result on pod number revealed that the plants treated with difenoconazole (0.1%) recorded the highest (360.00 pods/plot) number of pods while, the lowest number (180.66 pods/pot) was noticed in untreated control plants.

With regards to yield, the plants treated with difenoconazole (0.1%) registered the maximum (1620 kg/ha) yield while the untreated control plants registered the lowest yield (863kg/ha)

From the present study, it was concluded that foliar application of difenoconazole (0.1%) at an interval of 35, 50 and 65 DAS was found to be the most effective treatment in managing Cercospora leaf spot in vegetable cowpea besides supporting proportionately attractive pod yield (1620 kg/ha) at an acceptable B:C ratio (2.44:1).Among the organic formulations, mineral oil (0.1%) was found to be the effective treatment in controlling leaf spot with a pod yield of 1293 kg /ha .While, *P. fluorescens* recorded a pod yield of 1318 kg/ha with B: C ratio of 2.02:1

REFERENCES

REFERENCES

Abebe,G., Hattar, B. and Al-Tawaha, A. R. M. 2005. Nutrient availability as affected by manure application to cowpea (*Vigna unguiculata* .L. Walp) on calacarious soils. *J. Agric. Soc. Sci.* 1(1):1- 6.

Adegbite,A.A. and Amusa,N.A. 2008. The major economic field diseases of cowpea in the humid agro-ecologies of south-western Nigeria. *African J. biotechnology*: 7 (25): 4706-4712.

Agrios, G. N. 2005. *Plant Pathology,* Academic Press, UK, London, 922p.

Allen, D. J.,Thottappilly, G., Emechebe, A. M. and Singh, B. B. 1998. Diseases of cowpea. In: Allen, D. J. and Lenne, J. M. (eds) *The pathology of food and pasture Legumes*.CAB International, Wallingford, Oxon, UK. pp. 267-308.

Ali, M., Joshi, P. K., Pande, S.,Asokan, M ., Virmani, S. M. and Kandpal, B. K (2000) *Legumes in the Indo-Gangetic Plain of India.* In: Legumes in rice and wheat cropping systems of the Indo-Gangetic Plain - constraints and opportunities. International Crops Research Institute for the Semi-Arid Tropics, Patancheru, Andhra Pradesh, India, pp. 35-70.

Benagi, V. I . 1995. Epidemiology and management of late leaf spot of groundnut caused by *Phaeoisariopsis personata* Ph.D. *Thesis* University of Agricultural Sciences.Dharwad, 259p.

Bhat, F. A., Bhat, G. N., Anwar, A. and Mohiddin, F.A. 2015.Cost effective strategies for the management of fungal leaf spot of green gram caused by *Cercospora*

canescens Ell. & Mart. under temperate condition of Jammu and Kashmir. *Legume Res. 38*(1): 109-114.

Bird, J. and Maramorosch, K. 1975. *Tropical Diseases of Legumes.* Acad. Press, London. 171p.

Brown, M.C., Waller, C.D., Charlet, C. and Palmer, R.1986. The use of flutriafol based fungicides for the control of sugar beet diseases in Europe. Proceedings of the British Crop Protection Conference – Pests and Diseases. Farnham, UK: British Crop Protection Council, 1055 – 61.

Butler, E.J.1973. *Fungi and diseases in Plants*. Bishen Singh and periodicals experts. Delhi. 547pp.

Chowdhury, S. 1945. Control of *Cercospora* blight of tilt. *Indian J. Agric. Sci.*15: 140-142.

Chupp, C.1954. A Monograph of the fungus genus *Cercospora ithaca,* New York 667pp.

Crous, P. W. and Braun, U. 2003. Mycosphaerella and its anamorphs: 1. Names published in Cercospora and Passalora.Central bureauvoor Schimmel cultures, Utrecht, The Netherlands.

Das, S.R. and Mohanty, N.N. 1986. Chemical Control of Cercospora Leaf Spot of Green gram . *Indian Journal of Plant Protection*. 14(1):41- 43

Dahmen,H. and Stau,T.1992. Protective,curative and eradicant activity of difenoconazole against *Venturia inequalis*, *Cercospora arachdicola* and *Alternaria solani*. *Plant Dis*. 76:774-777.

Devappa,V. and Thejakumar, M.B.2016. Integrated management of chilli leaf spot caused by *Alternaria alternata* and *Cercospora capsici* under field conditions. International *J.* of Advanced Research. 4(4): 1468-1474.

Ellis, M. B. 1976.*More Dematiaceous Hyphomycetes.* CAB International Mycological Institute, Kew, U.K. 507p.

Emechebe, A.M. and Shoyinka, S. A. 1985. Fungal and bacterial diseases of cowpeas in Africa. In: Singh, S. R. and Rachie, K. O. (eds). *Cowpea: Research, Production and Utilization*. John Wiley, New York, pp.173-192.

Fery, R.L. and Dukes, P. D. 1977. Cercospora leaf spot of southern pea: studies on yield-loss and genetics of resistance. *Hortic. Sci.* 12(3):234.

Ghuge, S. S., Mayee C. D. and Godbole G. M. 1981. Assessment of losses in peanut due torust and tikka leaf spots. *Indian Phytopathol.* 34(2): 179-182.

Gill, H. S. and Singh. R.1962.Some *Cercospora* species from India – VII. *Indian Phytopathol.*17(2) : 243 – 246.

Hemachandra, H. 2007. Studies on leaf spot of tropical sugarbeet caused by *Cercospora beticola* Sacc. M. Sc. (Ag) Thesis, Tamil Nadu Agriculture University, Coimbatore, 95 p.

Hemachandra, H., Prakasam,V. and Jadesha, G. 2011.*In vitro* screening of fungicides and botanicals against *Cercospora beticola* Sacc. Paper presented in : *Nation.Symp. on Integrated Disease Management Strategies in Relation to Climate Change in South India, UAS Dharwad*, pp. 80-81.

Hennebert, G.L.and Sutton, B.C. 1994 .Unitary Parameters in Conidiogenesis. In: Ascomycete Systematics, Problems and Perspective in the Nineties(ed. DL Hawksworth), NATO ASI Series 296, New York, USA. 65–76.

Hundekar, A. R., Jahagirdar, S.,Matiwade, P. S. and Bhat, B. N. 2005. Management of frog eye leaf spot of bidi tobacco through chemicals. In: *National Conference on Tobacco*; 3-7 October, 2005, Hyderabad. Angrau, Hyderabad, 100 p.

Imtaij,A.,Rahman,S.A.,Parvin,R.,Farhana,K.M.,Kim,S.B.and Lee,T.S.2005.Effect of fungicidesmand plant extract on the conidial germination of *Colletotrichum gloeosporioides* causing mango anthracnose. *Mycobiol.*33(4):65-72.

Irena Gaurilcikiene, Irena Deveikyte and Egle Petraitiene, 2006, Epidemic progress of *Cercospora beticola* Sacc. in *Beta Vulgaris* under different conditions and cultivar resistance. *Biologia.*, 4:54-59.

Jahagirdar, S. and Hundekar, A. R. 2010. Management strategies against frog eye leaf spot pathogen (*Cercospora nicotianae*) in India. *Indian Phytopathol.* 63(1):63-65.

Jameel, A., Lal, H.C., Yogesh, K., Singh, P.K., Jyotirmoy, G., Zakauallah, K. and Gautam, N.K. 2014. Multiple disease resistance in greengram and blackgram germplasm and management through chemicals under rain-fed conditions *legume res.*37 (1): 101 – 109.

Kapadiya, H. J. and Dhruji, I. U.1999.Management of mungbean Cercospora leaf spot through fungicides. *Indian phytopathol.* 52(1):96-97.

Kaur, A., Gupta, V. P. and Singh, R. B. 2004. Chemotheraphy- an efficient tool to control Cercospora leaf spot in mungbean. *J. Mycol. Pl. Pathol.* 34:515-516.

KAU [Kerala Agricultural University].2009.*Package of Practices Recommendations (Adhoc) for organic farming:Crops*. Kerala Agriculture University,Thrissur,200p.

Khadar, S. K. 1999. Management of foliar diseases of groundnut with special reference to botanicals. M.Sc.(Ag) thesis, University of Agricultural Sciences, Dharwad, 82p.

Khader, S.K. 1999. Management of foliar diseases of groundnut (*Arachis hypogea* L.) with special reference to botanical.M.Sc. (Ag.) thesis. University of Agricultural Sciences, Dharwad, 85p.

Khan, M.I. and Rishi, K .1990. Antifungal activity of leaf extract of neem on seed mycoflora of wheat. *Indian J. Appl. And Pure Biol.* 5:131-134.

Kiraly, Z., Klement, S.J., Voros,H. and Solymosy, K. 1974. Methods in plant pathology with special reference to breeding for resistance. Elsevier Scientific Publishing Company, New York, 212p.

Kumar, P. and Mishra, U. S. 1992. Diseases of *Sesamum indicum* in Rohilkhand: intensity and yield loss. *Indian Phytopathol. 45*(1): 121-122.

Kumar, R., Manjul, P. and Ramesh, C. 2011. Effect of relative humidity, temperature and fungicide on germination of conidia of *Cercospora canescens* caused the Cercospora leaf spot disease in mungbean." *Archives of phytopathol and plant protection* .44(16): 1635-1645

Kumar, S., Jaiswal, S., Lal, A.A., Kumar, A. and Verma, A. 2017. Influence of natural products and bio-fungicide against tikka disease of groundnut caused by *Cercospora* spp.Pharma Innovation Journal; 6(3): 213-216.

Lang, L., Yu, Z., Zheng, Z., Xu, M., and Ying, H. 1993. Faba bean in China: state-of-the-art review. ICARDA, pp. 64-83.

Liu, L., Kloepper, J.W. and Tuzun, S. 1995. Induction of systemic resistance in cucumber against Fusarium wilt by plant growth-promoting rhizobacteria. *Phytopathol*, 85: 695-698.

Meghvansi, M. K., Khan, M. H., Gupta, R. and Veer, V.2013. Identification of a new species of *Cercospora* causing leaf spot disease in *Capsicum assamicum* in north eastern India. *Res Microbiol.*164(9):894-902.

Mulder, J.L. and Holliday, P.1975. C.M.I. Descriptions of pathogenic fungi and bacteria.463p.

Munjal, R. L., Lall, G. and Chona, B. 1960. Some Cercospora species from India IV. *Indian Phytopathol.* 13:144-145.

Oladiran, A.O. 1983.Studies on the disease of Cowpea (*Vigna unguiculata* (L.)Walp) Ph.D. thesis, University of Adan, Nigeria, 225 p.

Pairashi, M. 2007. Studies on frog eye leaf spot of bidi tobacco caused by *Cercospora nicotianae* Ell. & Eve. M. Sc. (Ag) Thesis, University of Agricultural Sciences, Dharwad,95 p.

Palakshappa, M. G., Parameswarappa, K. G. and Salimath, P. M. 2011. Management of Cercospora leaf spot of sesame (*Sesamum indicum* L.) caused by *Cercospora* spp.,National symposium on integrated disease management strategies in relation to climate change in South India, 30p.

Pant, D.C. 1989.Perpetuation of leaf spot organism of Lobiya. *Indian Phytopathol.* 42(1):187-188.

Pandey, K. K. and Pandey, P. K. 2002. Incidence of cowpea foliar blight caused by *Pseudocercospora cruenta* in relation to weather factors. *Indian Phytopathol.* 55 (2): 206-209.

Pandey, S., Sharma, M., Kumari, S., Gaur, P.M,, Chen, W., Kaur, L., Macleod, W., Basandrai, A.K., Basandrai, D., Bakr, A., Sandhu, J.S., Tripathi, H.S. and Gowda, C.L.L. 2009. Integrated foliar diseases management of legumes. In: Grain Legumes: Genetic improvement, Management and Trade. Indian Institute of Pulses Research, Kanpur, India .pp.143-161.

Patel, B. N., Patel, H. R., Patel, P. M., Bhatt, N. A., Patel, K. D., Patel, J. G.and Patel, S. J. 2001. Assessment of avoidable yield loss due to frog eye disease in bidi tobacco. *Tob. Res*. 27 (2): 187-189.

Pons, N., Sutton, B.C. and Gay, J.L. 1985. Ultrastructure of conidiogenesis in *Cercospora beticola*. *Trans. Br.Mycol. Soc*.85: 405-416.

Poornima. 2010. Studies on *Cercospora beticola* Sacc. causing leaf spot of palak (*Beta vulgaris* var.*bengalensis* Hort.).M.Sc. (Ag) thesis, University of Agricultural Sciences, Dharwad, 90 p.

Punja, Z.K, and Utkhede, R.S. 2003. Using fungi and yeasts to manage vegetable crop diseases. *Trends in Biotechnology*. 21: 400-407.

Puranik, S. B., Patil Kulkarni, B.G., Patil, N.K., Padaganur, G.M. and Hiremath, R. V. 1973. Fungicidal control of tikka disease of groundnut in Karnataka state. *Oilseeds J.* 3: 24-28.

Raguchander,T. and Samiyappan, K. P. R. 2005. The field evaluation of *Pseudomonas fluorescens* and *Bacillus subtiuson* the management of Cercospora leaf spot and powdery mildew in urdbean. *Legume Res*. 28(2): 137-139.

Ranga, R.G.V., Desai, S., Rupela, O.P, Krishnappa, K. and Wani, S.P. 2007. Integrated Pest Management Options for Better Crop Production. Procedings of the comprehensive Assessment of watershed programs in India. ICRISAT pp. 155-175.

Reddy, F. S. N., Nagarajan, K. and Ramjilal., R.1992.Effect of fungicides on control of frog eye spot disease (*Cercospora nicotianae)* of burley tobacco. *Tob. Res*., 18(1&2): 159-161.

Saxena, P. and Tripathi, H. S. 2006. Fungicidal management of Cercospora leaf spot of mung bean (*Vigna radiata*). *J. Mycol. Pl. Pathol*. 36(2): 336-337.

Saxena, M., Saxena, D.R., Bhale, M.S. and Khare,M.N. 1998. Diseases of cowpea and their management. In: *Diseases of Field Crops and their Management* (ed. Thind T.S.), National Agricultural Technology Information Centre, Ludhiana, India. pp 239-252.

Schwartz, H.F. and Pastor -Corrales, M.A. 1989. Bean production problems in the tropics, Cali, Colombia: CIA. 2p

Scheneider, R. W.,Williams, R. J. and Sinclair, J. B. 1976. Cercospora leaf spot of cowpea. Models for estimating yield loss. *Phytopathol.*66: 384-388.

Schneider,R.W. and Sinclair.1975.Inhiition of conidial germination and germ tube growth of *Cercospora canescens* by cowpea leaf diffusates.*Phytopathol* 65:63-65.

Simonetti, E., Carmona, M.A., Scandiani, M.M., Garc, A.F.,Luque, A. G., Correa, O.S. and Balestrasse, K.B.2012.Evaluation of indigenous bacterial strains for biocontrol of the frogeye leaf spot of soyabean caused by *Cercospora sojina*. The Society for Applied Microbiology Letters in Applied Microbiology 55, 170–173.

Singh, D.,Singh, N .,Gill, J. S. and Singh, P.2012 . Management of bacterial blight and leaf spot diseases of upland cotton using *Pseudomonas fluorescens*. Punjab Agricultural University, Regional Station, Faridkot , India. J. of Cotton Research and Development 26(1) :99-100.

Singh, R. A., De, R. K., Gurha, S. N. and Ghosh, A. 2001. Management of cercospora leaf spot of mungbean. In: National Symposium on pulses for Sustainable Agriculture and National Security, New Delhi, ISPRD, IIPR, Kanpur, India, pp 146.

Singh,S.N., Yadav.P., Sinha,S.K. and Ojha,K.L.1998.Efficacy of Plant Extract in Inhibition of Radial Growth of *Colletotrichum Capsici*. *J. of applied biology,*51:180-183.

Singh, S. P., Singh, S.K. and Shukla,V. 2013.Fungicidal Management of Cercospora Leaf Spot of Mungbean (*Vigna radiata*). 6(6): 861-863.

Singh, R. K. and Dwivedi, R. S. 1987. Effect of oils on *Sclerotiumn rolfsii* causing root rot of barley. *Ind. J. Phytopath.*, 40, 531-533.

Solheim,W.G.1929.Morphological studies of the genus *Cercospora* III. Monographs in population biology ,12:1–15.

Srivastava, S. and Nelson, S. 2012. Cercospora leaf spot of eggplant. *Plant Dis.*82:1-5.

Stover, R. 1990.Bananas, 3rdEdition, Longman Scientific and Technical, Harlow, Essez, p. 468.

Sundaram, N.V. (1965). Note on creation of tikka disease of groundnut. Indian Oilseeds J., 9: 98.

Sunkad, G. and Mesta, R. K.2005. Field efficacy of some fungicides for effective and economical control of major foliar diseases of groundnut. *Karnataka J. Agric. Sci. 18*(4): 995-997.

Swamy, K. M. 2010. Epidemiology and management of leaf spot of chilli (*Capsicum annuum* L.) caused by *Cercospora capsici.* M.Sc. (Ag) thesis, University of Agricultural Sciences, Raichur, 89 p.

Thammaiah, N. and Shirol, A. M. 2013. Management of sigatoka leaf spot disease of banana with oil based formulations.*Int. J. Agric. Sci. Vet. Med.*

Thammaih, N. 2003. Studies on epidemiology and management of Sigatoka leaf spot of banana. Ph. D. thesis, University of Agricultural Sciences, Dharwad, 85p.

Tiwari, A. and Kotastane, S. R.1984. Chemical control of fungal foliar diseases of mungbean. *Indian Phytopathol.* 37: 383.

Trivedi,A. 2014 .Management of viral and leaf spot disease complexes in organic farming of blackgram. *Indian Phytopathol* .67(1): 97-101.

Usha, C. and Rekha, P.G. 2014. Efficacy of leaf exudate of *Jatropha curcas* on percentage spore germination inhibition of its selected phylloplane and rhizosphere fungi .*Indian J. Sci.Res*. 4 (1): 70-74

Usman, M.R., Jaganathan, R. and Dinakaran, D. 1991. Plant disease management on groundnut with naturally occurring plant products. *Madras Agric. J.*78: 152-153.

Vaishnav, K. A, Patel, V. A., Dhedhi, B. M. and Kikani, B. R. 1992. Chemical management of leaf spot of sesamum caused by *Cercospra sesame* Zimm. *Indian Phytopathol.Abst. Papers*. p. 92

Vakli, N. G. 1977. Field screening of cowpea for Cercospora leaf spot resistance. *Trop. Agric*. 54: 69-76.

Vasudeva, R. S. 1963.*Indian Cercosporae*, Indian Council of Agricultural Research. Publication, p. 245.

Veena .,Yashoda, R., Hegde,A., Divyajyoti, U. and Priya, H. R. 2014.Survey and integrated management of fungal foliar diseases of green gram. *Environ. Life Sci.*7(3) :165- 168.

Verma, P. R. and Patel,P. N. 1969. Host range, varietal resistance and epidemiological observations of Cercospora leaf spot disease of cowpea. *Indian Phytopathol.*27: 61-66.

West, M. 1983. New systemic fungicide highly effective on spot and speckle, *banana Bull.* 47: 4-6.

Weinert, E. J.,Miller, S. A., Ikeda, D. M., Chang, K. C. S., McGinn, J. M. and DuPonte, M. W. 2014. Natural farming fish amino acid, sustainable

agriculture, M. Sc. (Ag.) thesis, University of Hawaii, College of Tropical Agriculture and Human Resources, Honolulu.104p.

Wheeler, B. E. J. 1969. An introduction to plant diseases. John Wiley and Sons Ltd., London.

Williams, R. J. 1975.Diseases of cowpea (*Vigna unguiculata* L.) Walp.) in Nigeria. Pest Articles & News Summaries.21: 253-267.

Williams, R. J. 1977.Identification of multiple disease resistance in cowpea. *Trop.Agric.* 54(1): 53-59.

Yu, T. F. 1947. Cercospora leaf spot of broad bean in China. *Phytopathol.* 37:174–179.

APPENDIX

APPENDIX

COMPOSITION OF STAIN USED

1. **Lactophenol –Cotton blue**

Anhydrous lactophenol	-67.00 mL
Distilled water	-20.00mL
Cotton blue	-0.10g

Anhydrous lactophenol prepared by dissolving 20 g phenol in 16ml lactic acid in 3ml glycerol.

ABSTRACT

MANAGEMENT OF CERCOSPORA LEAF SPOT OF VEGETABLE COWPEA (*Vigna unguiculata* subsp. *Sesquipedalis* (L.) Verdcourt.).

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Abstract of the thesis

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ABSTRACT

The study entitled "Management of Cercospora leaf spot of vegetable cowpea *Vigna unguiculata* subsp. *ungiculata (*L.) Verdcourt) was undertaken at the Department of Plant Pathology, College of Agriculture Vellayani and Coconut Research Station, Balaramapuram during 2015-2017 with the objective to study the symptomatology and etiology of Cercospora leaf spot of cowpea and to develop a management strategy.

Symptomatology and etiology of Cercospora leaf spot disease was studied during the flowering or pod bearing stage of the crop from different locations near to College of Agriculture, Vellayani. Symptoms were observed on the leaves, pods and stem .Conidial dimension of the pathogen ranges from 24.64 to 57.6 µm x 1.06 to 2.7 µm and the conidial septation was around 10 to 18. Based on the conidial and mycelial characters, the pathogen was tentatively identified as *Cercospora* sp. The identity of the pathogen was further confirmed based on the morphological characteristics and the herbarium specimen send to National Fungal Culture Collection of India, (N.F.C.C.I), Pune as *Pseudocercospora* sp. aff. *Pseudocercospora vignigena* (F: Mycosphaerellaceae)

In vitro pathogen suppression by spore germination assay revealed that the fungicide difenoconazole (0.1%) recorded 42.96 per cent reduction over control and was followed by propiconazole (0.1%) and hexaconazole (0.1%) which recorded 36.85 and 31.10 per cent reduction over control, respectively. The organic formulations, mineral oil (0.1%) and neem oil (0.5%) recorded only 21.88 per cent reduction over control and were statistically on par .KAU talc based formulation of *Pseudomonas fluorescens* at 2.0 % recorded 11.88 per cent reduction of spore germination over control.

The pot culture studies conducted using eight best treatments (difenoconazole (0.1%), propiconazole (0.1%), hexaconazole (0.1%), carbendazim (0.05%), azoxystrobin (0.05%), mineral oil (0.1%) neem oil (0.5%) and *P. fluorescens* 2.0 %) selected from *in vitro* studies revealed that the foliar spray of 0.1 per cent difenoconazole recorded the minimum disease incidence (23.80%) and disease severity (8.23 %) that accounts to 54.80 and 76.06 per cent disease reduction over control respectively. In the case of organic formulations, mineral oil (0.1%) recorded the minimum disease incidence (30.04%) and disease severity (16.54%). The plant sprayed with KAU talc based formulation of *P. fluorescens* (2.0%) recorded 33.69 % disease severity reduction over control. With regard to pod yield, the plants treated with difenoconazole (0.1%) registered the maximum (0.523 kg/pot) yield with 88.80% yield increase over control while ,the untreated control plants registered the lowest yield (0.277kg/pot).

Based on the results of pot experiment, a field study was conducted at Coconut Research Station, Balaramapuram to evaluate the efficacy of five best treatments (difenoconazole (0.1%), propiconazole (0.1%), mineral oil (0.1%) neem oil (0.5%) and *P. fluorescens* 2.0 %) selected from the pot culture studies. Among the fungicides, difenoconazole (0.1%) recorded the minimum (31.88%) disease incidence and disease severity (13.97%) compared to all other treatments. In the case of organic formulations, mineral oil (0.1%) gave the maximum (14.32%) disease suppression and the lowest disease incidence of 40. 57 %. With regard to yield, maximum pod yield was from the plot sprayed with difenoconazole (1620 kg /ha) followed by propiconazole (1386.5 kg /ha) treated plants. Even though, the application of *P. fluorescens* gave only 29.23 per cent of the disease suppression, it enhanced the yield by 52.27 per cent yield over the control.

It was concluded that foliar application of difenoconazole (0.1%) at an interval of 35, 50 and 65 DAS was found to be the most effective treatment in managing Cercospora leaf spot in vegetable cowpea besides supporting

proportionately attractive pod yield (1620 kg/ha) at an acceptable B:C ratio (2.44:1).Among the organic formulations, mineral oil (0.1%) was found to be the effective treatment in controlling leaf spot with a pod yield of 1293 kg/ha . While, *P. fluorescens* recorded a pod yield of 1318 kg/ha with B: C ratio of 2.02:1